JPRS 70633

14 February 1978

USSR

# SPACE BIOLOGY AND AEROSPACE MEDICINE

Vol. 11, No. 6, 1977





U. S. JOINT PUBLICATIONS RESEARCH SERVICE



142

NOTE

JPRS publications contain information primarily from foreign newspapers, periodicals and books, but also from news agency transmissions and broadcasts. Materials from foreign-language sources are translated; those from English-language sources are transcribed or reprinted, with the original phrasing and other characteristics retained.

Headlines, editorial reports, and material enclosed in brackets [] are supplied by JPRS. Processing indicators such as [Text] or [Excerpt] in the first line of each item, or following the last line of a brief, indicate how the original information was processed. Where no processing indicator is given, the information was summarized or extracted.

Unfamiliar names rendered phonetically or transliterated are enclosed in parentheses. Words or names preceded by a question mark and enclosed in parentheses were not clear in the original but have been supplied as appropriate in context. Other unattributed parenthetical notes within the body of an item originate with the source. Times within items are as given by source.

The contents of this publication in no way represent the policies, views or attitudes of the U.S. Government.

PROCUREMENT OF PUBLICATIONS

JPRS publications may be ordered from the National Technical Information Service (NTIS), Springfield, Virginia 22161. In ordering, it is recommended that the JPRS number, title, date and author, if applicable, of publication be cited.

Current JPRS publications are announced in Government Reports Announcements issued semimonthly by the NTIS, and are listed in the Monthly Catalog of U.S. Government Publications issued by the Superintendent of Documents, U.S. Government Printing Office, Washington, D.C. 20402.

Correspondence pertaining to matters other than procurement may be addressed to Joint Publications Research Service, 1000 North Glebe Road, Arlington, Virginia 22201.

| BIBLIOGRAPHIC DATA SHEET | 1. Report No. JPRS 70633 | 2. | 3. Recipient's Accession No. |
|---|---|---|---|
| 4. Title and Subtitle<br>SPACE BIOLOGY AND AEROSPACE MEDICINE, Vol.11 , No.6, 1977 | | | 5. Report Date<br>14 February 1978<br>6. |
| 7. Author(s) | | | 8. Performing Organization Rept. No. |
| 9. Performing Organization Name and Address<br>Joint Publications Research Service<br>1000 North Glebe Road<br>Arlington, Virginia 22201 | | | 10. Project/Task/Work Unit No.<br>11. Contract/Grant No. |
| 12. Sponsoring Organization Name and Address<br>As above | | | 13. Type of Report & Period Covered<br>14. |

15. Supplementary Notes
Cover-to-cover translation of KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA, bimonthly journal, published by the USSR Ministry of Health.

16. Abstracts

The report contains articles concerning the selection and training of cosmonauts; evaluation and analysis of accumulated data to facilitate the on-going transition from orbital to interplanetary flights; research aimed at guaranteeing safety on long flights and reliability of the human component of the "man-spaceship" system; space psychology and physiology; environmental problems and control (spacecraft habitability, effects of radiation and weightlessness, etc.) and telemetry.

17. Key Words and Document Analysis. 17a. Descriptors

USSR
Exobiology
Life Support
Human Factors Engineering
Confined Environments
Stress (Psychology)
Aerospace Environment
Biotelemetry

17b. Identifiers/Open-Ended Terms

17c. COSATI Field/Group 6S, 6K

| 18. Availability Statement | 19. Security Class (This Report) | 21. No. of Pages |
|---|---|---|
| Unlimited Availability<br>Sold by NTIS<br>Springfield, Virginia 22151 | UNCLASSIFIED | 143 |
| | 20. Security Class (This Page) UNCLASSIFIED | 22. Price A07 |

JPRS 70633

14 February 1978

# SPACE BIOLOGY AND AEROSPACE MEDICINE

Vol. 11, No. 6, 1977

Complete translation of the Russian-language periodical KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA published in Moscow by the Meditsina Izdatel'stvo

## CONTENTS

CONTENTS (Continued) Page

CONTENTS (Continued) Page

PUBLICATION DATA


| | |
|---|---|
| English title | : Space Biology and Aerospace Medicine |
| Russian title | : Kosmicheskaya Biologiya i Aviakosmicheskaya Meditsina |
| Author (s) | : |
| Editor (s) | : O. G. Gazenko |
| Publishing House | : Meditsina |
| Place of Publication | : Chekhov, Moscow Oblast |
| Date of Publication | : November-December 1977 |
| Signed to press | : 13 October 1977 |
| Copies | : 1703 |
| COPYRIGHT | : Kosmicheskaya biologiya i aviakosmicheskaya meditsina, 1977 |

EXPERIMENTAL AND GENERAL THEORETICAL RESEARCH

UDC: 612.11-057:629.786.2"Salyut-4"

# CHANGES IN PERIPHERAL BLOOD OF CREW MEMBERS OF THE SALYUT-4 ORBITAL STATION

Moscow KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA in Russian
No 6, 1977 pp 3-12

[Article by V. I. Legen'kov, R. K. Kiselev, V. I. Gudim and G. P. Moskaleva,
submitted 12 Dec 75]

[Text] It is known that the medullary type of hemopoiesis in mammals is phylogenetically determined by the need to maintain a high level of metabolism and increasing oxygen requirement under gravitational conditions. A decrease in intensity of physical loads and absence of gravity should diminish hemopoiesis [1, 2].

Modern manned space flights on orbital stations take place with prolonged exposure to weightlessness and relative hypodynamia [3, 4]. As we know, the study of hematological changes induced by long-term space flights is one of the pressing problems of space biology and medicine.

In this work, we submit the hematological indices of crew members of the Salyut-4 orbital station and, for the sake of comparison, the same parameters of the crew of Salyut-3 after a 16-day mission.

## Methods

We used the method of G. V. Derviz and A. I. Vorob'yeva[5] in the modification of I. M. Babkova and S. S. Vedenskiy [6] to assay peripheral blood hemoglobin. The conventional methods used in clinical practice [7, 8] served to assay erythrocytes and leukocytes per microliter blood, as well as to calculate the Price-Jones curve and determine, on celloscopes 101 and 302, the amount of thrombocytes per microliter blood, the leukocyte formula and reticulocytogram. We counted the absolute number of eosinophils by the method of Dunger [9] in the modification of I. S. Piralishvili [10], and the absolute number of basophils by the methods of Moore and James [11], Braunsteiner and Thumb [12]. Total hemoglobin mass was determined by the carbon monoxide method based on the specific capacity of carbon monoxide to bind with blood hemoglobin [13]. Erythropoietin was examined on polycythemic hypoxic mice by the method of Cotes and Bangham as modified by T. I. Koretskaya et al. [14]. We evaluated erythropoietic activity according to the number of reticulocytes; saline and standard erythropoietic C, calibrated by the international B standard, served as a control [15].

Table 1. Dynamics of some hematological indices in crew members of Salyut-3 and Salyut-4 orbital stations before, during and after 16-, 30- and 63-day missions (n-2)

| Index | 16-day mission | | | | | | | 30-day mission | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | before flight | | during flight | | after flight | | | before | | after flight | | | |
| | day of examination | | | | | | | day of examination | | | | | |
| | 75 | 6 | 5—9 | 15 | 0 | 1 | 5 | 58—44 | 7 | 0 | 1 | 3 | 7 |
| Hemoglobin, g% | 14,54 | 14,83 | — | — | 15,05 | 14,90 | 14,46 | 14,38 | 14,69 | 15,35 | 15,00 | 13,50 | 13,17 |
| Erythr.,million/μl | 4,825 | 4,935 | — | — | 5,310 | 5,095 | 5,210 | 5,000 | 4,805 | 3,9435 | 4,065 | 4,220 | 4,1375 |
| Color index | 0,904 | 0,900 | — | — | 0,850 | 0,894 | 0,834 | 0,870 | 0,919 | 1,180 | 1,125 | 0,960 | 0,963 |
| Mean diameter of erythr.,μm | — | 7,5 | — | — | — | — | 8,2 | 7,5 | — | — | — | — | — |
| Osmotic resist. of erythr., min | 0,475 | | | | | | 0,450 | 0,450 | | | | | |
| max | 0,360 | — | — | — | — | — | 0,350 | 0,350 | — | — | — | — | — |
| Acid resist.of erythrocytes | Negative | — | — | — | — | — | Negat. | Negat. | — | — | — | Negat. | — |
| Reticulocytes per μl | 33 775 | 39 480 | — | — | 26 550 | 22 927,5 | 44 285 | 39 550 | 40 550 | 30 118,5 | 34 732,5 | 54 830 | 100 937,5 |
| Reticulocytes per 1000 erythrocytes | 7,0 | 8,0 | — | 5,6 | 5,0 | 4,5 | 8,5 | 7,99 | 8,4 | 7,6 | 8,5 | 13,04 | 26,8 |
| Reticulocytes per μl: | | | | | | | | | | | | | |
| skeins | 0 | — | — | — | — | — | — | — | 0 | — | — | — | 0 |
| clumped | 388,1 | — | — | — | — | — | 0 | — | 0 | — | — | — | 0 |
| net-)complete | 2715,6 | — | — | — | — | — | 4265,4 | — | 5068,75 | — | — | — | 36 842,2 |
| work)incomplete | 12 497,1 | — | — | — | — | — | 13 098,75 | — | 12 976,0 | — | — | — | 38 356,25 |
| granules | 18 174,2 | — | — | — | — | — | 27 431,15 | — | 22 505,25 | — | — | — | 25 739,06 |
| Thrombocytes per μl | 251 275 | 224 805 | — | — | 263 205 | 190 050 | 226 175 | 218 000 | 299 500 | 238440 | 223 741 | 253 800 | 371 200 |
| Thrombocytes per 1000 erythr. | 52,0 | 45,5 | 50,5 | 47,0 | 49,5 | 37,5 | 43,5 | 43,6 | 62,3 | 65,0 | 55,0 | 60,1 | 89,7 |
| Blood-clotting time, s | 525,0 | 462,5 | — | — | 358,5 | — | 523,5 | 540 | 445 | 382 | — | 425 | — |
| Bleeding time,s | 75,0 | 142,5 | — | — | 52,5 | — | 105,0 | 85 | 138 | 125,5 | — | 182,5 | — |
| Hematocrit, % | 44,0 | 44,5 | — | — | 50,25 | 46,75 | 43,5 | 44,5 | 45 | 42,5 | 42,5 | — | — |
| ESR, mm/h | 2,0 | 2,5 | — | — | 2,0 | 3,0 | 4,0 | 3,5 | 3,0 | 3,0 | 4,5 | 4,5 | 3,0 |

Table 1, continued

| Index | 30-day mission | | | 63-day mission | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | after flight | | | before flight | | | flight | after flight | | | | |
| | day of examination | | | day of examination | | | | | | | | |
| | 12 | 30 | 45 | 94 | 26 | 3 | 51 | 0 | 2 | 7 | 16 | 39—45 |
| Hemoglobin, g% | 13,10 | 14,42 | 14,68 | 15,52 | 15,16 | 15,27 | — | 14,90 | 13,72 | 13,40 | 14,60 | 15,22 |
| Erythr., million/μl | 4,615 | 5,020 | 4,740 | 4,970 | 5,200 | 5,110 | — | 4,450 | 3 900 | 4,230 | 4,820 | 5,190 |
| Color index | 0,854 | 0,860 | 0,937 | 0,970 | 0,874 | 0,898 | — | 1,01 | 1,055 | 0,950 | 0,910 | 0,890 |
| Mean erythroc. diameter, μm | 8,2 | — | 7,5 | 7,5 | — | — | — | — | 6,1 | 6,1 | 6,8 | 7,15 |
| Osmotic resist. of erythr., min / max | 0,475 / 0,380 | — | — | 0,450 / 0,350 | — | — | — | — | 0,50 / 0,34 | 0,42 / 0,35 | 0,43 / 0,31 | — |
| Acid resist.of erythrocytes | — | — | — | Negat. | — | — | — | — | Negat. | — | — | — |
| Reticulocytes per μl | 104 255 | 57 605 | 58 610 | 27 360 | 34 160 | 38 385 | — | 15 645 | 27 335 | 54 920 | 88 750 | 59 941 |
| Reticulocytes per 1000 erythrocytes | 22,5 | 11,47 | 12,3 | 5,4 | 6,57 | 7,5 | 4,0 | 3,5 | 7,0 | 13,0 | 18,5 | 11,5 |
| Reticulocytes per μl: | | | | | | | | | | | | |
| skeins | — | — | — | — | 0 | — | 0 | 0 | — | 0 | 0 | 0 |
| clumps | — | — | — | — | 0 | — | 0 | 0 | — | 2471,4 | 2662,5 | 1198,8 |
| net-)complete | — | — | — | — | 4270,0 | — | 14,0 | 2503,3 | — | 9885,6 | 21 300,0 | 11 988,2 |
| works)incompl. | — | — | — | — | 14 151,6 | — | 30,0 | 5006,4 | — | 19 771,2 | 32 837,5 | 22 477,9 |
| granules | — | — | — | — | 16 396,8 | — | 56,0 | 8135,4 | — | 22 791,8 | 31 950,0 | 24 276,1 |
| Thrombocytes per μl | 241 460 | 293 045 | 196 850 | 231 330 | 199 840 | 288 155 | — | 174 110 | 218 820 | 187 850 | 196 320 | 286 685 |
| Thrombocytes per 1000 erythrocytes | 52,3 | 58,3 | 41,6 | 46,6 | 38,45 | 56,3 | 42 | 39,0 | 56,0 | 44,50 | 45,5 | 55,5 |
| Blood-clotting time, s | — | — | — | 457,5 | — | 515 | — | — | 313,5 | 507,5 | 525,5 | — |
| Bleed.time, s | — | — | — | 107,5 | — | 140 | — | — | 87,5 | 70,0 | 114,5 | — |
| Hematocrit, % | — | 44,5 | 44,75 | — | 37,75 | 40,5 | — | 33,75 | 35,50 | 35,50 | 35,75 | 38,0 |
| ESR, mm/h | 4,0 | 3,0 | 3,5 | 6,0 | 4,5 | 5,5 | — | 9,0 | 8,0 | 4,5 | 5,5 | 5,0 |

Note: Mean values are given here and in Table 2.

Table 2. Dynamics of leukocytogram of crew members on Salyut-3 and Salyut-4 orbital stations, before, during and after 16-, 30- and 63-day missions (n = 2)

| Index | 16-day mission | | | | | | | 30-day mission | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | before | | during | | after flight | | | before | | after flight | | | | |
| | day of examination | | | | | | | day of examination | | | | | | |
| | 75 | 6 | 5–9 | 15 | 0 | 1 | 5 | 58–44 | 7 | 0 | 1 | 3 | 7 | 12 |
| Leukocytes per μl | 8000 | 6825 | — | — | 9850 | 8250 | 7550 | 6750 | 8400 | 8250 | 7150 | 6250 | 6450 | 6750 |
| Leukocytes per 1000 erythrocytes | 1,65 | 1,38 | 1,485 | 1,09 | 1,85 | 1,61 | 1,44 | 1,35 | 1,74 | 2,09 | 1,75 | 1,48 | 1,56 | 1,46 |
| Leukocytogram:* myelocytes | 0 / 0 | 0 / 0 | 0 / 0 | 0 / 0 | 0 / 0 | 0 / 0 | 0 / 0 | 0 / 0 | 0 / 0 | 0 / 0 | 0 / 0 | 0 / 0 | 0 / 0 | 0 / 0 |
| metamyelocytes | 0 / 0 | 0 / 0 | 0 / 0 | 0 / 0 | 0 / 0 | 0 / 0 | 0 / 0 | 0 / 0 | 0 / 0 | 0 / 0 | 0 / 0 | 0 / 0 | 0 / 0 | 0 / 0 |
| stab nuclear neutrophils | 3,77 / 301,5 | 3,19 / 218,0 | 3,13 / — | 2,13 / — | 6,11 / 602,5 | 5,5 / 454,0 | 2,0 / 151,0 | 1,5 / 95,5 | 3,5 / 294,5 | 3,0 / 247,5 | 3,75 / 254,25 | 4,5 / 292,5 | 3,75 / 243,25 | 3,5 / 252,5 |
| segmented neutrophils | 57,14 / 4571,0 | 55,74 / 3804,0 | 62,17 / — | 62,0 / — | 64,45 / 6349,0 | 56,0 / 4681,0 | 53,44 / 4035,0 | 54,75 / 3761,5 | 52,25 / 3883,25 | 66,0 / 5468,0 | 61,0 / 4343,0 | 47,75 / 2990,0 | 49,0 / 3163,25 | 51,0 / 3685,25 |
| basophils | 0,25 / 19,5 | 0,3 / 20,0 | 0,52 / — | 0,0 / — | 0,5 / 49,0 | 1,0 / 82,5 | 0,4 / 39,0 | 0,25 / 19,75 | 0,5 / 42,0 | 0,25 / 27,25 | 0,5 / 35,75 | 0,75 / 51,5 | 0,75 / 49,75 | 0,5 / 32,75 |
| eosinophils | 2,0 / 160,0 | 1,1 / 75,0 | 4,7 / — | 2,0 / — | 0,26 / 26,0 | 2,00 / 165,0 | 2,48 / 187,5 | 2,25 / 160,5 | 2,75 / 391,25 | 1,25 / 109,75 | 1,0 / 80,75 | 3,0 / 210,0 | 2,0 / 134,5 | 5,0 / 272,5 |
| lymphocytes | 28,85 / 2308,0 | 32,78 / 2237,5 | 28,72 / — | 29,13 / — | 23,44 / 2307,0 | 23,76 / 1960,5 | 30,0 / 2266,0 | 32,5 / 2101,5 | 36,0 / 3029,0 | 23,5 / 1904,0 | 28,5 / 2057,0 | 37,5 / 2287,5 | 33,0 / 2079,0 | 33,5 / 2017,5 |
| monocytes | 8,0 / 640,0 | 6,89 / 470,5 | 5,22 / — | 5,41 / — | 5,44 / 535,6 | 11,0 / 907,0 | 11,66 / 880,45 | 8,75 / 610,75 | 5,0 / 420,0 | 6,0 / 468,5 | 5,5 / 411,75 | 6,5 / 417,5 | 6,5 / 430,25 | 7,5 / 520,75 |
| Absolute number of eosinophils per μl | 93,5 | 28,3 | — | — | 3,125 | 0,0 | 196,5 | 146,75 | 19,5 | 47,0 | 65,5 | 267,5 | — | — |
| Absolute number of basophils per μl | 3,0 | — | — | — | 14,0 | 14,0 | 4,45 | 15,4 | 17,15 | 3,1 | 14,05 | — | — | — |

Table 2, continued

| Index | 30-day mission after flight | | 63-day mission | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | day of examin. | | before flight | | | during flight | | | | after flight | | | |
| | | | day of examination | | | | | | | | | | |
| | 30 | 45 | 94 | 26 | 3 | 5 | 30 | 45 | 51 | 0 | 2 | 7 | 16 | 39—45 |
| Leukocytes per μl | 7700 | 5950 | 5850 | 7800 | 6125 | — | — | — | — | 12900 | 6950 | 6350 | 7450 | 6675 |
| Leukocytes per 1000 erythrocytes | 1,53 | 1,21 | 1,18 | 1,5 | 1,2 | 1,5 | 1,35 | 1,10 | 1,01 | 2,89 | 1,78 | 1,51 | 1,54 | 1,30 |
| Leukocytogram:* | | | | | | | | | | | | | | |
| myelocytes | 0/0 | 0/0 | 0/0 | 0/0 | 0/0 | 0,0 | 0,0 | 0,0 | 0,0 | 0/0 | 0/0 | 0/0 | 0/0 | 0/0 |
| metamyelocytes | 0/0 | 0/0 | 0/0 | 0/0 | 0/0 | 0,0 | 0,0 | 0,0 | 0,0 | 0,5/68,25 | 0,5/49,5 | 0/0 | 0,5/38,0 | 0/0 |
| stab nuclear neutrophils | 2,5/205 | 2,0/110,75 | 3,75/224,25 | 3,5/273,0 | 3,5/214,37 | 2,5 | 1,5 | 2,75 | 1,75 | 10,25/1346,62 | 4,0/307,5 | 3,75/251,7 | 6,5/485,0 | 3,0/200,25 |
| segmented neutrophils | 51,5/4152,0 | 52,5/3189,75 | 57,0/3276,0 | 59,25/4767,75 | 58,25/3556,87 | 58,0 | 50,5 | 50,0 | 52,0 | 71,0/9087,75 | 58,5/3992 | 58,0/3644,25 | 54,75/4067,25 | 54,5/3637,9 |
| basophils | 1,0/77,0 | 0,25/19,0 | 0,25/13,0 | 0,25/22,75 | 0,5/30,62 | 0,0 | 0,0 | 0,0 | 0,0 | 0/0 | 0,75/44,75 | 0,5/31,7 | 1,0/73,75 | 0,5/33,4 |
| eosinophils | 1,0/77,0 | 1,75/100,0 | 2,75/169,0 | 2,0/143,0 | 2,75/170,0 | 1,5 | 2,0 | 3,0 | 2,5 | 0,25/34,12 | 2,25/178,5 | 2,75/188,17 | 3,25/244,75 | 2,0/133,5 |
| lymphocytes | 35,0/2470,0 | 36,5/2339,25 | 30,75/1839,75 | 29,25/2161,25 | 29,0/1779,37 | 30,0 | 41,5 | 40,0 | 37,75 | 12,25/1627,12 | 26,25/1876 | 27,75/1779,55 | 30,0/2078,25 | 31,5/2102,6 |
| monocytes | 9,0/718,0 | 7,0/441,25 | 5,5/328,25 | 5,75/432,25 | 6,0/373,75 | 8,0 | 4,5 | 4,25 | 6,0 | 5,75/736,12 | 8,25/501,25 | 7,25/454,55 | 6,25/463,0 | 6,0/480,5 |
| Absolute eosinophils per μl | 140,0 | 165,6 | 115,5 | 172,0 | 169,0 | — | — | — | — | 28,12 | 128,12 | 252,25 | 165,6 | 99,9 |
| Absolute basophils per μl | 6,25 | 7,25 | 7,77 | 15,75 | 21,5 | — | — | — | — | 3,0 | 1,55 | 12,5 | 17,2 | 7,82 |

*Percentile content is given in the numerator and number of cells per microliter, in the denominator.

Table 3. Changes in hemoglobin mass in crew members of Salyut-3 and Salyut-4 orbital stations

| | Duration of mission, days | Before flight | | | | After flight, day of examination | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | date of examination | hemoglobin | date of examin. | hemoglobin | 5 | 12 | 16 | 30 | 45 |
| Salyut-3: | | | | | | | | | | |
| P.R. Popovich | 16 | 15/II 1973 | 825 / 435 | 19/IV 1974 | 780 / 410 | 660 / 348 | — | — | — | — |
| Yu. P. Artyukhin | 16 | » | 792 / 460 | » | 790 / 461 | 715 / 415 | — | — | — | — |
| Salyut-4, first crew: | | | | | | | | | | |
| A. A. Gubarev | 30 | 11/VII 1972 | 722 / 388 | 29/XI 1974 | 710 / 380 | — | 537 / 290 | | 682 / 368 | 760 / 410 |
| G.M.Grechko | 30 | » | 720 / 396 | 15/XI 1974 | 695 / 380 | — | 495 / 270 | — | 520 / 284 | 630 / 344 |
| Second crew: | | | | | | | | | | |
| P.I.Klimuk | 63 | 14/VII 1972 | 645 / 380 | 15/XI 1974 | 622 / 360 | — | — | 520 / 301 | — | — |
| V. I. Sevast'yanov | 63 | » | 590 / 346 | » | 635 / 370 | — | — | 475 / 276 | — | 608 / 349 |

Note: Hemoglobin mass (in grams) is given in the numerator and hemoglobin mass ($g/m^2$ body surface), in the denominator.

Table 4. Erythropoietic activity of cosmonaut blood and urine before and after mission (according to data on polycythemic hypoxic mice)

| Time of examination, day | Base data | | Duration of mission, days | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | | 16 | | 30 | | 63 | |
| | polycythemic mouse reticulocytes (0/00) | | | | | | | |
| | blood | urine | blood | urine | blood | urine | blood | urine |
| Before flight | 4,4±0,6 (25) | 0,8±0,1 (5) | | | | | | |
| After flight: | | | 16,0±2,6* (14) | 3,2±1,4 (4) | 24,6±3,5* (7) | 8,6±1,4 (12) | — | 4,5±1,6 (8) |
| 0-day | | | | | | | | |
| 2d day | | | | | | | 13,8±3,4** (6) | 3,7±0,9 (7) |
| 4th | | | 12,7±4,1*** (6) | 2,3±1,1 (3) | | | | |
| 5th | | | 4,0±1,8 (2) | 4,0±1,1 (4) | | | | |
| 7th | | | | | 18,6±1,1* (11) | 4,8±1,5 (9) | 10,5±2,2** (4) | 5,6±1,3 (8) |
| 30th | | | | | 24,1±1,1* (9) | — | | |
| 39th | | | | | | | | |
| 45th | | | | | 3,0±0,9 (7) | | 8,2±2,9 (5) | |
| G standard | 15,0±0,6 (20) | | 16,4±1,0 (10) | | 15,1±1,1 (9) | | 13,2±1,6 (4) | |
| Saline | 2,5±0,3 (20) | | 2,3±1,0 (10) | | 1,8±1,0 (10) | | 1,8±0,8 (4) | |

Note: P was calculated in relation to base data:
*--P = 0.001 **--P = 0.01 ***--P = 0.05

The number of experimental mice is shown in parentheses.

Results and Discussion

Preflight diagnostic-laboratory and clinical examination of the crew members of orbital space stations failed to demonstrate any pathological changes in the blood system.

In the prelift-off period, there was a decline of blood eosinophils to 70-87% and a tendency toward increased excretion of hormones in urine, which is apparently related to nervous and emotional tension.

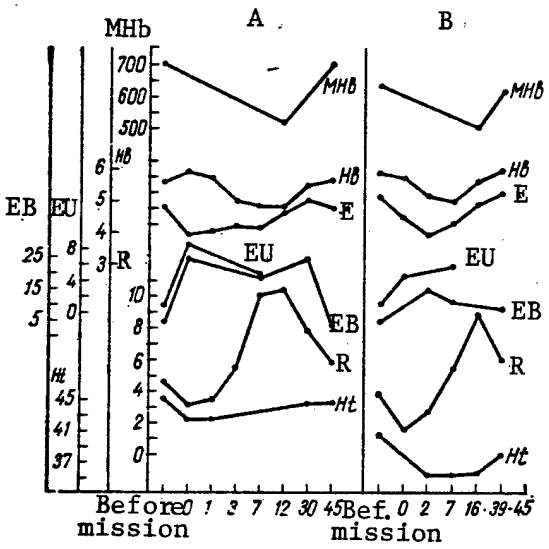
During and after the missions, the cosmonauts presented similar changes occurring in the same direction. Analysis of the data submitted in Tables 1-4 shows that the reticulocyte level, as well as total number of leukocytes, were low during the mission on Salyut-3 and in the second crew of Salyut-4. In the leukocyte formula, there was a decline of all morphological elements of bone marrow hemopoiesis.

The concentration of hemoglobin in peripheral blood was somewhat lower in the first few hours after landing than during the periods of prelift-off and preflight examination, after the 16- and 30-day missions. In the second crew (63-day mission), there was some decrease in hemoglobin after landing, as compared to preflight data. However, the color index on zero day was somewhat higher than the background indices after the 30- and 63-day missions (0.915 and 0.898 before the mission, 1.180 and 1.01 after). Perhaps this is attributable to changes in morphology of erythrocytes, i.e., presence in blood of both normal erythrocytes and spherocytes. Evidently, this hyperchromatism is due to an increase in thickness of erythrocytes. Microscopy revealed that some of the erythrocytes were of a darker color and the central lumen was considerably reduced, i.e., light passed through the thicker layer of hemoglobin.

Soon after landing (after 4-4.5 h), all of the cosmonauts presented 34% less reticulocytes than the background levels (26,106±4581 and 39.472±622, respectively). The erythropoietin level in blood taken 5-18 h after landing was 3-5 times higher than the base levels in the members of the three crews, and there was also an elevation of urine erythropoietin level on 0 day. In the next 7-12 days of the readaptation period, the first and second crews of Salyut-4 presented a mean 15.2% decline of erythrocytes, 11.5% decrease in concentration of hemoglobin, 11% decrease in hematocrit and 23.6±3.0% decrease in total hemoglobin mass (see Figure). At this time and thereafter (until the blood indices were restored), the erythropoietin level remained above the base values. The elevation of erythropoietin level demonstrated on the 1st postflight day was associated with reticulocytosis, which was an indication of activation of erythropoiesis.

Thus, in the case of the 30-day mission, the number of reticulocytes began to increase gradually and, by the 7th-12th postflight days, it increased by almost 3.5 times, with a significant increase in reticulocytes of grades III and IV maturity on the reticulocytogram [16]. Analogous

changes were noted in the second crew. However, the reticulocytogram showed a more marked shift to the left and appearance of younger forms (to grade II). With increase in amount of reticulocytes, there was a gradual increase in erythrocytes, concentration of hemoglobin, total hemoglobin mass and hematocrit index.




Dynamics of some hematological indices of the first (A) and second (B) crews of Salyut-4 before and after the missions.

X-axis, days before mission, 0 day and after mission; y-axis, indices:
Total hemoglobin in body (MHb, g)
Hemoglobin in peripheral blood (Hb, g%)
Erythrocytes per microliter blood ($E \cdot 10^6$)
Erythropoietic activity of urine (EU, %)
Erythropoietic activity of blood (EB, %)
Reticulocytes per μl blood ($R \cdot 10^4$)
Hematocrit (Ht, %)

The fact that, concurrently with the changes in erythropoietin level and other blood indices, there was a left shift of the Price-Jones curve in the second crew of Salyut-4 merits attention. The curves of erythrocyte diameters showed an increase in microcytes, and the mean diameter constituted 6.1 μm. Normalization of the Price-Jones curve occurred by the 39th-45th postflight day. In addition, some of the indices used in examination of erythrocytes (mean thickness of an individual erythrocyte, 3.21 μm; index of spherical shape, 1.9; mean area of erythrocyte, 116.80 $\mu m^2$) also presented a shift in the direction of microcytosis by the 7th postflight day.

The brief increase in total number of leukocytes, which was observed at the time of landing and which returned to its original level 24-48 h after the space mission, evidently occurs as a result of a high level of hormones in blood and urine, and it is due to partial redistribution of blood and the stress effect of all space factors on the body at the final stage of the mission, i.e., landing. This is confirmed by the transient eosinophilopenia, which reverts to base data 2-3, or 5-7 days at the most after landing, when the hormone concentration drops. In the crew of Salyut-4, the thrombocyte content dropped by a mean of 30% and was gradually restored by the 7th-16th postflight day. Perhaps this is related to the general inhibition of hemopoiesis in weightlessness, and in particular of thrombocytopoiesis.

The observed decrease in number of reticulocytes in flight and the presence of mild signs of normochromic anemia for 1 month after the mission indicate that changes appear in biosynthetic processes under weightless and relatively hypodynamic conditions [3, 17].

Perhaps the decrease in reticulocytes is indicative of destruction thereof in the spleen or depression of erythropoiesis. The latter hypothesis is more realistic, since restoration of blood erythrocytes takes place for 1 month or longer.

We cannot rule out the possibility that the gradually developing hyperbaric hypoxia on space stations [3, 4] has a deleterious effect on blood, which is related to formation of peroxide compounds [18-22], as well as, apparently, substances that inhibit erythropoiesis.

On 0 day, there was a sharp elevation of blood erythropoietic titer to 0.24-0.5 units/ml. Perhaps intensification of production thereof is related to anemic hypoxia of tissues at the start of the readaptation period and subsequent activation of redox processes in the organism.

Long-term model experiments conducted on earth, in which flight diets were used and all space factors, with the exception of weightlessness, were simulated, revealed that there were no changes in erythrocytes. There was no change in hemoglobin mass in 30- and 90-day experiments. This suggests that weightlessness is the prime space factor leading to changes in the red blood system.

The nature of restoration of blood indices in the readaptation period is indicative of an inconsistency between restoration of reticulocyte content and changes in indices of hematocrit, concentration of erythrocytes and hemoglobin. Against the background of reticulocytosis, these indices remained low for a long time (particularly in the 30-day mission), very probably because there was faster restoration of circulating blood volume than erythrocyte mass and hemoglobin synthesis.

Thus, in weightless and with relative hypodynamia, as well as in the presence of other space factors, the human body undergoes specific stages of adaptation to the new environment, which is reflected in the cell composition of peripheral blood and concentration of hemoglobin. Apparently, the new quantitative and qualitative blood indices are more optimum under such conditions.

The shifts and changes in blood demonstrated in the cosmonauts after the missions should apparently be interpreted as a certain base condition when changing from weightlessness to earth's gravity, which, on the whole, leads to increased production of erythropoietin, which stimulates erythropoiesis.

BIBLIOGRAPHY


1. Korzhuyev, P. A. in: "Aviatsionnaya i kosmicheskaya meditsina" [Aviation and Space Medicine], Moscow, 1963, pp 284-287.

2. Idem, "Hemoglobin," Moscow, 1964.

3. Legen'kov, V. I. "Dynamics of Peripheral Blood Indices of Cosmonauts in the Course of Occupational Training and During Space Missions," candidatorial dissertation, Moscow, 1974.

4. Gurovskiy, N. N.; Yeremin, A. V.; Gazenko, O. G.; et al. KOSMICHESKAYA BIOL. [Space Biology], No 2, 1975, pp 48-54.

5. Derviz, G. V., and Vorob'yeva, A. I. LABOR. DELO [Laboratory Record], No 3, 1959, pp 3-8.

6. Babkova, I. M., and Vedenskiy, S. S. Ibid, No 9, 1966, p 562.

7. Predtechenskiy, V. Ye. "Manual of Clinical Laboratory Tests," Moscow, 1960.

8. Todorov, Y. "Clinical Laboratory Tests in Pediatrics," Sofia, 1963.

9. Dunger, R. MUNCH. MED. WSCHR., Vol 89, 1942, p 57.

10. Piralishvili, I. S. LABOR. DELO, No 3, 1962, pp 20-22.

11. Moore, J. E., and James, Y. W. PROC. SOC. EXP. BIOL. (New York), Vol 82, 1958, p 60.

12. Braunsteiner, H., and Thumb, H. ACTA HAEMAT. (Basel), Vol 20, 1958, p 339.

13. Kiselev, R. K. "A New Carbon Monoxide Technique for Assaying Hemoglobin," candidatorial dissertation, Moscow, 1973.

14. Karetskaya, T. I.; Moskaleva, G. P.; Gudim, V. I.; et al. PAT. FIZIOL. [Pathological Physiology], No 3, 1969, pp 77-79.

15. Gudin, V. I.; Moskaleva, G. P.; Koretskaya, T. I.; et al. PROBL. GEMATOL. [Problems of Hematology], No 10, 1971, pp 48-49.

16. Kassirskiy, I. A., and Alekseyev, G. A. "Clinical Hematology," Moscow, 1955, pp 61-89.

17. Makarov, G. A. in: "Molekulyarnyye aspekty regulyatsii eritropoeza" [Molecular Aspects of Regulation of Erythropoiesis], Ryazan', 1974, pp 132-139.

18. Jensen, W. N. ASTRONAUT. ACTA, Vol 17, 1972, pp 223-227.

19. Shmeleva, A. M. in: "Problemy kosmicheskoy meditsiny" [Problems of Space Medicine], Moscow, 1966, pp 388-389.

20. Zhironkin, A. G.; Breslav, I. S.; et al. in: "Problemy kosmicheskoy biologii" [Problems of Space Biology], Moscow, Vol 4, Pt 3, 1965, pp 518-530.

21. Keisall, M. A. UNIV. COLORADO STUDIES, SER. BIOL., No 4, 1958, p 62.

22. Yeliseyev, V. G. "Connective Tissue," Moscow, 1961.

UDC: 612.014.477-063

# HUMAN ENDURANCE OF RECURRENT $+G_z$ ACCELERATIONS

Moscow KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA in Russian
No 6, 1977 pp 12-19

[Article by S. R. Kotovskaya, R. A. Vartbaronov and M. N. Khomenko,
submitted 5 Jan 77]

[Text] We know from the data in the literature that maximum accelerations in flight could reach 8-9 units, and in a number of instances they exceed the pilot's physiological capabilities, even if anti-G gear is used [1-3]. The most adverse consequences of exposure to $+G_z$ in flight are manifested in the form of visual disturbances, fainting, cardiac and respiratory disorders; after difficult missions there is increased fatigability and appearance of some pathological changes in the organism [2, 4, 5].

Most of these adverse effects have been studied very comprehensively under laboratory conditions, but only in the case of single exposure to this factor. Yet the dynamics of change in inflight accelerations are characterized by recurrent "peaks" of $+G_z$, which differ in magnitude, duration and acceleration gradient. In this regard, it may be assumed that recurrent exposure imposes a certain imprint on the distinctions of a pilot's endurance of various modes of inflight accelerations. We could expect development of both cumulative and adaptational effects [6-12].

The objective of this work was to investigate endurance of recurrent $+G_z$ accelerations that are closer to piloting conditions. For this, it was necessary to study the following: 1) maximum tolerance of recurrent accelerations as related to acceleration gradient, as well as magnitude of maximum and minimum accelerations; 2) distinctions and dynamics of physiological reactions in the course of recurrent exposure to accelerations; 3) effect of operator activity on endurance of recurrent accelerations.

## Methods

Six males, 19-21 years of age, deemed physically fit for flight work, with no restrictions, participated in our studies. All of the studies were made on a centrifuge with a radius of 7.25 m. The angle of inclination of the chair back constituted 17° back from the vertical position.

At the first stage, we tested maximum endurance of single exposure to head-pelvis $+G_z$ accelerations, using the conventional method of expert evaluation of pilot endurance on a centrifuge [3]. Mean stability of the subjects, as determined by this method, constituted 6.5±0.7 units.

It should be noted that, according to the data of P. M. Suvorov, mean endurance of pilots under analogous conditions constitutes 5.84±0.8 units [13]. Consequently, the subjects we tested can be equated with a group of healthy pilots with rather high endurance of accelerations.

We conducted 44 tests in 7 series to test endurance of recurrent exposure to accelerations.

Rotation on the centrifuge was effected in accordance with a program with automatic generation of various modes of peak-like* accelerations of up to 8.5 U [units] with a linear acceleration gradient. In all instances, the subjects deliberately effected prelum abdominale and tensed the muscles of the lower extremities at accelerations of 4 U or more.

In all of the studies, we recorded the dynamics of change in accelerations, EKG in the three leads according to Neb, systolic arterial pressure in the auricular concha, minute volume of respiration, EMG of the abdominal rectus and femoral quadriceps muscles. The appearance of the subject was monitored continuously and there was continuous communication with him through a radiotelevision system.

In the five main series of studies, we varied the magnitude of maximum, minimum and moderate integral accelerations, as well as the acceleration gradient. Repeated exposure lasted until the first signs of decompensation appeared. If none was present, the entire exposure period did not exceed 11 min.

In all instances, the regular anti-G suit was worn with recurrent exposure to accelerations.

Taking into consideration the scatter of parameters of G-forces, which was expected, according to our data [14], under actual flight conditions, we used 2 levels for each of them: 1) magnitude of maximum peak G-force (7.5 and 8.5 U); 2) magnitude of minimum G-force (1.2 and 3 U); 3) duration of one cycle (16 and 32 s).

The total number of possible combinations (series) constitutes $m^n$, where $m$ is the number of levels and $n$ is the number of factors (in our case, $2^3 = 8$). We selected five of all the possible number of series (series II-VI).

*Continuous exposure to maximum level of each "peak" of acceleration lasted 1 s.

In addition, in the first series of studies, additional operator demands were made of the subject. For this purpose, we used the method of simple [unidimensional] compensatory monitoring of a luminous marker, deflections of which on the oscilloscope screen were proportionate to the magnitude of existing G-forces. The regular control lever was used to compensate for the deflections.

In series VII, we simulated peaks of recurrent G-forces of 5.5/1.5 U with an increment gradient of 0.4 U/s and standard duration of exposure of 6 min. In this series, the choice of acceleration parameters was made on the basis of mean levels thereof obtained under actual flight conditions [14].

Table 1 lists the modes of recurrent accelerations and distribution of subjects in different series.

Table 1. Modes of recurrent exposure to accelerations and distribution of subjects

| Series | G-force magnitude, U | | | Cycle of exposure, s | Gradient, U/s | Number | | Operator activity involved | Maximum exposure time, min |
|---|---|---|---|---|---|---|---|---|---|
| | maxim. | minim. | mean | | | examin. | cases | | |
| I | 7,5 | 1,2 | 4,35 | 32 | 0,39 | 5 | 5 | Yes | 10,7 |
| II | 7,5 | 1,2 | 4,35 | 32 | 0,39 | 5 | 6 | No | 8,8 |
| III | 7,5 | 1,2 | 4,35 | 16 | 0,78 | 5 | 6 | " | 10,1 |
| IV | 7,5 | 3,0 | 5,25 | 16 | 0,56 | 6 | 7 | " | 5,5 |
| V | 7,5 | 3,0 | 5,25 | 32 | 0,28 | 3 | 5 | " | 4,6 |
| VI | 8,5 | 3,0 | 5,75 | 16 | 0,69 | 5 | 5 | " | 4,5 |
| VII | 5,5 | 1,5 | 3,5 | 22 | 0,36 | 5 | 10 | " | 6,0 |
| Totals | — | — | — | — | — | 6 | 44 | — | — |
| Extreme values | 5,5—8,5 | 1,2—3,0 | 3,5—5,75 | 16—32 | 0,28—0,78 | — | — | — | 4,5—10,7 |

In order to neutralize the conditioning factor, we varied the order in which a subject was used in different series of studies, with the exception of the 5th and 7th series, which were conducted after the preceding ones were terminated.

Results and Discussion

The lack of data in the literature concerning maximum endurance of recurrent exposure to G-forces in the head-pelvis direction compelled us to pay attention to the distinctive manifestations of subjective disorders inherent in the modes we tested. Visual disorders, which occurred in 50% of the

cases (Table 2), were the most frequent finding limiting endurance of G-forces. It is important to note that visual disturbances, in the form of a "gray veil," usually appeared after repeating several cycles of exposure and only at the peak of 7.5 or 8.5 U G-force. When the tests were continued the visual disturbances increased to the appearance of a "black veil" (after 2-3 peaks), in spite of the fact that the subjects contracted their muscles somewhat more.

We must stress the brief and transient nature of the visual disorders, which always disappeared with lowering of G-forces. These disorders were absent in the 7th series of studies, where maximum acceleration did not exceed 5.5 U.

The demonstrated cumulative effects of recurrent exposure to $+G_z$ were also observed according to other subjective indices. Thus, in 15% of the cases, the subjects' complaints of vertigo, which developed after a few cycles of exposure during periods close to the minimum G-force level, were the cause of discontinuing the tests. Analysis of the set of physical forces arising with rotation of the centrifuge revealed that this factor is related to the effects of maximum linear, angular and Coriolis accelerations, subjectively manifested in the form of the "rocking" phenomenon [5]. This phenomenon was particularly distinct when the cabin was significantly rotated (with a minimal radial component) to the original position and back, against the background of a revolving centrifuge. Consequently, the vestibular genesis of these sensations is unquestionable.

Finally, in isolated cases, there were complaints of breathing difficulty and pain in muscles of the legs after several cycles of exposure.

The deferred (cumulative) nature of the subjective symptoms was also confirmed, in some cases, by corresponding changes in some of the objective parameters (lowering of arterial pressure in the vessels of the concha and appearance of marked disturbances of cardiac rhythm).

Investigation of the critical signs enabled us to calculate the maximum time of endurance of various modes of recurrent peaks of head-pelvis accelerations (see Table 2). The group differences were evaluated by the method of paired comparison of series differing from one another primarily with regard to one of the above-mentioned factors (see Table 2 and Figure 1).

The greatest differences were found with reference to the factor of minimal G-force value, the role of which was evaluated by comparing the data for the 3d and 4th, or 2d and 5th series. It was found that an increase in level of minimal G-force from 1.2 to 3 U, with negligible change in gradient, leads to a 50% or greater reduction of mean endurance time.

A comparison of the results of other series (II and III, or IV and V), where the role of the acceleration gradient was evaluated, shows that a 2-fold increase thereof leads to a 25% increase in endurance of recurrent peaks of acceleration. Analogously, a comparison of the data for the 1st and 2d series demonstrates the significance of active operator work, which also increases acceleration endurance by about 30%.

Table 2. Maximum human endurance of various modes of recurrent peaks of $+G_z$ forces

| Series | Number of cases | Magnitude of G-f. units | | Duration of cycle, s | Operator activity | Subjects | | | | | | | | | | | | Mean endurance time in series, min (M±m) | Number of cases |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | | G-k | | S. | | K. | | L. | | R. | | G-y | | | |
| | | maximum | minimum | | | maximum endur., min | criterion of endur. | maximum endurance, minutes | criterion of endur. | maximum endurance, minutes | criterion of endur. | maximum endurance, min | criterion of endur. | maximum endurance, min | criterion of endur. | maximum endur., min | criterion of endur. | | |
| I | 5 | 7,5 | 1,2 | 32 | Yes | 8,8 | GV | >10,7 | — | >10,1 | — | >10,7 | — | >10,7 | — | — | — | >10,2±0,37 | 5 |
| II | 6 | 7,5 | 1,2 | 32 | No | 7,6 | GV | 8,8 | V | 7,6 | MP | 8,6 | GV | 7,0 | GV | — | — | 7,9±0,28 | 6 |
| III | 6 | 7,5 | 1,2 | 16 | No | 7,5 | V | 9,1 | V | >10,1 | — | >9,9 | — | >9,9 | — | — | — | >9,4±0,42 | 6 |
| IV | 7 | 7,5 | 3,0 | 16 | No | 3,6 | GV | 5,5 | GV, V | >4,7 | — | 5,0 | GV | 5,0 | GV | 4,6 | GV | 4,7±0,23 | 7 |
| V | 5 | 7,5 | 3,0 | 32 | No | — | — | — | — | 4,2 | V | 4,6 | GV | — | — | 3,6 | GV | 3,8±0,23 | 5 |
| | | | | | | | | | | 3,7 | RD | 3,2 | Es | | | | | | |
| VI | 5 | 8,5 | 3,0 | 16 | No | 3,1 | | 3,5 | GV | 3,1 | GV | 4,5 | GV | 4,2 | GV | — | — | 3,7±0,29 | 5 |

Notes: 1. Subject G-k in series II and subject K in series III and V of the studies were exposed to accelerations twice with the same results.

2. Criteria of maximum endurance:
GV--gray veil V--vertigo MP--muscular pain RD--respiratory difficulty
Es--extrasystoly >--limit not reached

3. Comparison of pairs revealed statistically unreliable differences between the means for series I and III. In all other cases the differences were reliable.

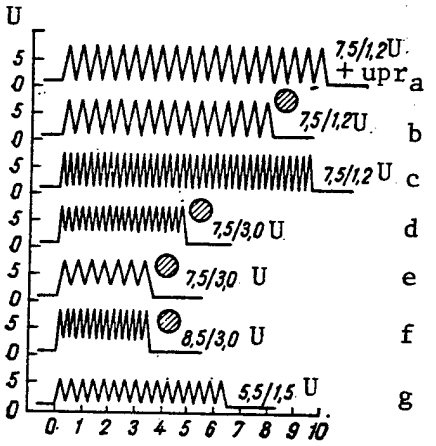



Figure 1.
Human endurance time for peak accelerations with exposure in different modes. X-axis, exposure time (min); y-axis, magnitude of $+G_z$ (units)
a) series I d) series IV g) series VII
b) series II e) series V
c) series III f) series VI
The numbers on the right refer to maximum and minimum G-forces in each series; lined circles refer to series in which maximum endurance was reached. Duration of one cycle of exposure: 16 s in series III, IV and VI, 22 s in series VII and 32 s in the others. [upr--expansion unknown]

A comparison of the results of the 4th and 6th series revealed a substantial level of maximum G-force, an increase in which of 1 U led to 21% decrease in endurance time.

Thus, the submitted results are indicative of importance of all four factors, various combinations of which were used in our series of investigations.

The results obtained in the studies of maximum endurance of recurrent exposure to peak accelerations can be approximately described by the following mathematical function: $t = \dot{G}^{0.33} \cdot 2^{7-75G_i}$, where $t$ is mean maximum endurance time in minutes; $\dot{G}$ is mean acceleration change gradient in U/s; $G_i$ is mean value of G-force, which equals half the sum of maximum and minimum values thereof.

It should be noted that, in a number of cases, development of cumulative effects was preceded by distinct manifestation of signs of compensatory and adaptational reactions. This was manifested, in some cases and more often in the 6th series, where maximum accelerations reached 8.5 U, by the fact that appearance of the "gray veil" in the first cycle of exposure was followed by total restoration of clear vision, already in the 2d "peak" of acceleration. This general manifestation of adaptation was studied in the 7th series of studies, which dealt with the distinctive features of development of physiological reactions to recurrent "peaks" of $+G_z$ constituting 5.5 U (Figure 2).

As can be seen in Figure 2, the pulse amplitude of the ear lobe (top curve), which had dropped in the 1st "peak" of acceleration, rapidly recovered starting in the 2d cycle and reached base levels by the end of the 2d min of exposure (4th-5th "peaks"). Concurrently there was rapid relative decline of bioelectrical activity of muscles and stabilization of tachycardia with accelerations in excess of 4 U. On the basis of the existing classification [16, 17], these phenomena can be attributed to development of the phase of incomplete (unstable) compensation. Starting with the end of the 2d min of

exposure (which corresponds to the 5th-6th "peak"), all of the indices are stabilized at one level, which is indicative of development of the second phase, the phase of complete compensation.

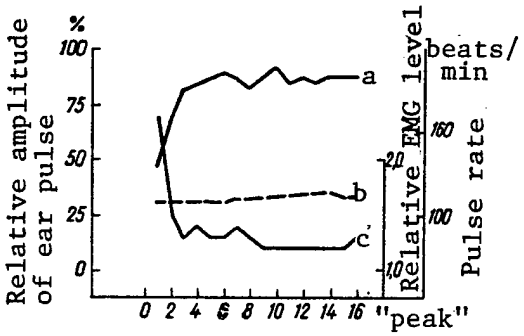



Figure 2.
Dynamics of physiological reactions of man to recurrent exposure to peak accelerations of 5.5/1.5 U lasting a total of 6 min.

a) relative amplitude of ear pulse (% of initial level)
b) mean pulse rate
c) relative bioelectrical activity of femoral quadriceps

Appearance of the third phase, the phase of progressive decompensation of physiological mechanisms, is inherent in repeated exposure to high accelerations ($+G_z$ = 7.5-8.5 U). The time of appearance of the third phase in different series of tests was related to the parameters of accelerations and severity of physical changes. The comparative results are submitted in Figure 3.

As can be seen in Figure 3, the results of the first 3 series of studies were characterized by relatively long phase of compensation and high arterial pressure in vessels of the ear lobe, moderate tachycardia and an increase in pulmonary ventilation.

The EMG of the femoral quadriceps muscle revealed, as in the 7th series, distinct adaptation starting already in the 2d min of exposure.

In the 4th and 6th series, there was a decline of arterial pressure in the vessels of the ear lobe, intensification of tachycardia and some increase in pulmonary ventilation, whichwas indicative of more intensive function of physiological systems. Unlike the preceding series of studies, there was faster progression of physiological changes inherent in depletion of compensatory capabilities of the organism. In particular, the rate of increase in mean pulse rate constituted 6-8/min, which is apparently an important prognostic criterion of subsequent decompensation.

The results of the 5th series, where systolic arterial pressure held at a rather high level (110 mm Hg), while endurance time was at a minimum as a result of the greater minimal acceleration (3.0 U) and minor gradient (0.3 U/s), merit special attention.

The cause of this inconsistency lies apparently in the fact that, in the 5th series, the participants were (as indicated above) quite conditioned to repeated exposure to $+G_z$ and they developed a higher level of muscular

tension (by about 30% as compared to the EMG level in the 1st "peak" of acceleration) and pulmonary ventilation (by more than 4 times).

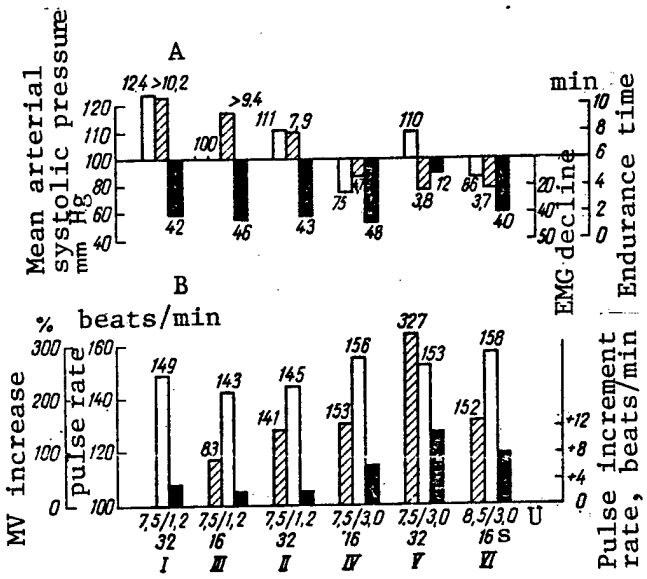



Figure 3. Distinctions of subjects' physiological reactions in different series of tests with repeated exposure to peak accelerations

A) white columns, mean arterial systolic pressure in the ear; striped, mean endurance time; black, relative EMG decline
B) white columns, mean pulse rate; striped, relative increment of minute volume of respiration (MV); black, rate of pulse increment
The Roman numerals refer to series of tests.

A typical finding in all series of tests was the total absence of correlation between tachycardia and acceleration level during the second exposure (with the exception of the period of acceleration build-up in the first cycle), which is consistent with the findings of inflight studies [18].

The submitted results indicate that the pattern of physiological changes was specific in each of the series of studies. Quantitatively, we found a correlation between severity of the changes and time of maximum endurance of recurrent accelerations. In particular, in most cases, when endurance time diminishes (series IV and VI) there was an increase in intensiveness of function of physiological systems and lowering of arterial pressure in vessels of the ear lobe. Only the very high tension of compensatory mechanisms (series V) permitted retention of a high level of circulation, although it could not prevent decompensation in the 3d min of exposure.

Thus, the above data are indicative of the substantial importance of the factor of recurrence of exposure in evaluating pilot endurance of G-forces occurring in flight. With a high intensity of piloting activity (mean accelerations of 5 U or more with a maximum in excess of 7 U), one should expect development primarily of cumulative effects of repeated exposure, and with lower levels there should be adaptational manifestations. For this reason, continued investigation of these questions, particularly under flight conditions, is a rather promising and pressing task.

## BIBLIOGRAPHY


1. Belostostkiy, Ye. M.; Popov, A. P.; and Samukhin, N. V. VOYEN.-SAN. DELO [Military Medical Record], No 7, 1940, p 5.

2. Borshchevskiy, I. Ya.; Mirolyubov, G. P.; Rozenblyum, D. Ye.; et al. Ibid, No 7, 1938, pp 11-16.

3. Armstrong, H. G., and Heim, J. W. J. AVIATION MED., Vol 9, 1938, pp 199-215.

4. Andreyev, V.; Appolonov, A.; and Sobennikov, I. VOYEN.-SAN. DELO, No 5-6, 1930, pp 51-57.

5. Sergeyev, A. A. "Physiological Mechanisms of Effects of Accelerations," Leningrad, 1967.

6. Babushkin, V. I.; Malkin, V. B.; and Usachev, V. V. VOYEN.-MED. ZH. [Military Medical Journal], No 4, 1956, pp 10-19.

7. Bobrovnitskiy, P. S.; Zagryadskiy, V. P.; Komendantov, G. L.; et al. in: "Voprosy aviatsionnoy meditsiny v GVF" [Problems of Aviation Medicine in the Civil Air Fleet], Moscow, 1962, pp 65-112.

8. Isakov, P. K. VOYEN.-MED. ZH., No 6, 1957, pp 65-72.

9. Savin, B. V. "Effects of G-Forces on Functional State of the Central Nervous System and Mechanism of Impairment of Its Function," author abstract of doctoral dissertation, Leningrad, 1966.

10. Flekkel', A. B., and Odinov, A. M. VOYEN.-MED. ZH., No 2, 1949, p 41.

11. Khazen, I. M.; Kogan, E. M.; and Barer, A. S. in: "Aviatsionnaya i kosmicheskaya meditsina" [Aviation and Space Medicine], Moscow, 1963, pp 469-472.

12. Wood, E. H.; Lambert, E. H.; and Code, C. F. J. AVIATION MED., Vol 18, 1947, pp 471-482.

13. Suvorov, P. M. "Physiological Studies Using the Centrifuge in the Practice of Medical Flight Expertise and Screening System," doctoral dissertation, Moscow, 1969.

14. Kolesnikov, S. V.; Vartbaronov, R. A.; and Kotovskaya, A. R. KOSMICHESKAYA BIOL. [Space Biology], No 4, 1976, pp 30-35.

15. Yuganov, Ye. M. in: "Problemy kosmicheskoy biologii" [Problems of Space Biology], Moscow, Vol 3, 1965, pp 54-69.

16. Barer, A. S. in: "Tezisy nauchnykh soobshcheniy 10-go s"yezda Vsesoyuznogo fiziologicheskogo o-va im. I. P. Pavlova" [Summaries of Scientific Papers Delivered at the 10th Congress of the All-Union Physiological Society imeni I. P. Pavlov], Moscow--Leningrad, Vol 2, Vyp 1, 1964, pp 77-78.

17. Rozenblyum, D. Ye. in: "Aviatsionnaya i kosmicheskaya meditsina," Moscow, 1963, pp 425-426.

18. Pircher, L. SCHWEIZ. MED. WSCHR., Vol 102, 1972. pp 444-447.

UDC: 612.74.015.348-06:612.014.477-064

# EFFECT OF PROLONGED WEIGHTLESSNESS ON METABOLISM OF PROTEINS IN RED AND WHITE SKELETAL MUSCLES OF RATS

Moscow KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA in Russian
No 6, 1977 pp 19-23

[Article by V. A. Kazaryan, E. A. Rapoport, L. A. Goncharova and S. Ya. Bulycheva, submitted 16 Jan 76]

[Text] It is known that prolonged exposure of higher animals and man to space flight conditions leads to a decrease in muscular strength and physical fitness, which could be partially related to development of atrophy of skeletal muscles, unless special preventive measures are taken [1]. It is important to define the nature of metabolic disturbances referable to muscular proteins in order to analyze the pathogenesis of functional deviations and atrophic reaction of muscles in weightlessness. We made a study, in this aspect, of biosynthesis of protein molecules in different structural components of muscles of the "fast" and "slow" type in rats after they had made a 22-day flight on the Kosmos-605 biosatellite. We compared the findings to data obtained at comparable times on animals involved in a concurrent [synchronous] ground-based experiment, in a spacecraft cabin with the same life support systems (LSS).

## Methods

Experiments were performed on four groups of Wistar male rats, with five animals in each group: the 1st and 3d groups consisted of "flight" rats on the 2d and 26th days, respectively, after landing; the 2d and 4th groups consisted of rats in the synchronous experiment on the 2d and 26th days, respectively, after removal from LSS. There were four groups of intact animals maintained in the vivarium on the same diet that served as controls. They were used in the experiments concurrently with the above-mentioned rat groups.

All of the animals were given intraperitoneal injections of 2-$^{14}$C-leucine at the rate of 5 μCi/250 g. One hour later they were given pentothal anesthesia and then exsanguinated by puncturing the vena cava, after which the leg muscles, m. soleus and m. extensor digitorum longus, were extracted and frozen in liquid nitrogen, and stored at -20° until they were treated.

We extracted protein fractions from the muscles using the system of I. I. Ivanov et al. [2]. We assayed them by the method of Lowry [3]. The proteins were submitted to the usual washing procedure [4] to prepare them from measurement of radioactivity, which was made with accuracy of 1-2% using a liquid scintillation SL-30 spectrometer and Bray's fluid as the scintillation mixture. We evaluated the results using the criterion of Student. The calculations were made on the Multi-20 computer using a program prepared in the LEM-S language.

Results and Discussion

As can be seen in Table 1, there was marked atrophy of the soleus (red) muscle in "flight" rats on the 2d postflight day. This was not demonstrable in the "synchronous" animals on the 2d day after LSS, in spite of relative hypokinesia. The weight of the red muscle was restored 25 days after the flight. Table 1 shows that on the 2d day after returning to the ordinary living conditions, some atrophy of the digital extensor longus (white muscle) was demonstrable in both "flight" and "synchronous" rats. At later stages, its weight in the rat groups we used did not differ from control levels. The demonstrated atrophy of both muscles examined was associated with virtually uniform decrease in amounts of sarcoplasmic, myofibrillar and stromal protein fractions, without change in percentile content thereof, if we do not count some relative elevation of stromal protein level in the atrophied red muscle. Restoration of muscle weight by the 26th day also failed to be associated with appreciable changes in fraction composition of proteins.

Table 1. Weight characteristics of red and white skeletal muscles of intact, "flight" and "synchronous" rats used in radioisotope experiments (M±m)

| Animal group | Rat weight, g | Muscle weight, mg | |
|---|---|---|---|
| | | red (m. soleus) | white (m. extensor d. l.) |
| "Flight," 2d day | 246±7 | 43±6* | 87±4* |
| Control | 279±21 | 68±7 | 106±6 |
| "Synchronous," 2d day | 266±12 | 74±5 | 83±8* |
| Control | 293±9 | 78±3 | 104±4 |
| "Flight," 26th day | 304±7 | 103±5 | 118±6 |
| Control | 301±11 | 94±5 | 123±5 |
| "Synchron.," 26th d. | 378±5* | 109±8 | 132±9 |
| Control | 327±16 | 118±4 | 130±6 |

Note: Here and in Tables 2 and 3, the asterisk indicates that the differences are reliable ($P<0.05$). The means of 5 readings for experimental rats (with the exception of the "synchronous" 2d group, from which 2 red muscles were taken) and 6-8 readings on control rats are shown here and in Table 2.

Table 2. Protein biosynthesis in red skeletal muscle of intact, "flight" and synchronous" rats (M±m)

| Animal group | Protein fraction | | | |
|---|---|---|---|---|
| | sarco-plasm | acto-myosin | T | stroma |
| | Specific radioactivity (counts/min/mg protein) | | | |
| "Flight," 2d day | 99±10 | 97±3* | 107±6* | 54±4* |
| Control | 77±4 | 78±2 | 72±2 | 43±2 |
| "Synchronous," 2d day | 120±9 | 103±1 | 79±1* | 105±0,4 |
| Control | 124±3 | 103±9 | 96±3 | 104±1 |
| "Flight," 26th day | 107±7 | 99±11 | 98±6 | 110±11 |
| Control | 99±3 | 93±5 | 96±3 | 105±5 |
| "Synchronous," 26th day | 80±9 | 84±3 | 79±4 | 89±2 |
| Control | 95±3 | 85±2 | 84±3 | 84±3 |
| | Total radioactivity (counts/min/muscle | | | |
| "Flight," 2d day | 300±40 | 276±25 | 214±18 | 153±16 |
| Control | 356±27 | 306±27 | 216±17 | 149±17 |
| "Synchronous," 2d day | 383±28 | 415±11 | 194±52 | 343±28 |
| Control | 429±13 | 403±20 | 253±18 | 365±14 |
| "Flight," 26th day | 535±52 | 574±57* | 549±43 | 564±66 |
| Control | 484±27 | 421±28 | 517±23 | 552±42 |
| "Synchronous," 26th day | 478±67 | 721±71 | 483±36 | 388±56 |
| Control | 589±32 | 725±38 | 526±19 | 457±38 |

Analysis of intensity of red muscle protein biosynthesis (Table 2) revealed that, in the "flight" rats, there was a shift in the same direction of specific radioactivity of all fractions on the 2d postflight day, i.e., an increase which was statistically significant with reference to actomyosin and the T fraction. However, when calculated for the entire muscle, incorporation of $^{14}$C-leucine in the same proteins did not differ from the findings in control vivarium animals. Hence, it can be concluded that overall synthesis of protein molecules was not appreciably altered in the atrophied soleus muscle. The increase in specific radioactivity of proteins in this muscle could have been due to lower dilution of newly formed labeled protein molecules by the suply of preformed unlabeled ones, the level of which was low. On the 26th day, total radioactivity of actomyosin of red muscle was somewhat higher than in the control, which was indicative of moderate prevalence of overall synthesis of this protein fraction toward the end of the period of readaptation to conditions on earth. We failed to demonstrate substantial changes in this muscle in "synchronous" rats, with respect to protein biosynthesis; there was merely minor depression of synthesis of components of the T fraction on the 2d day.

As can be seen in Table 3, incorporation of $^{14}$C-leucine in proteins of white muscle, on the 2d postflight day, was lower than incorporation in the same proteins of vivarium animals, with the exception of the sarcoplasmic fraction (converted per unit weight). The radioactivity of actomyosin and

fraction T per muscle was also reliably lower. All this was indicative of inhibited biosynthesis of these proteins. In "synchronous" animals, unlike the "flight" ones, on the 2d day after removal from LSS the intensity of protein synthesis in white muscle did not undergo appreciable change: there being some decline of specific radioactivity of stromal proteins, no reliable difference was observed between levels of overall radioactivity of the fractions examined and the same levels in control animals. At the later postflight stages, as well as post-LSS stages, no changes were demonstrated in protein biosynthesis in white muscle.

Table 3. Protein biosynthesis in white skeletal muscle of intact, "flight" and "synchronous" rats (M±m)

| Animal group | Protein fraction | | | |
|---|---|---|---|---|
| | sarcoplasm | actomyosin | T | stroma |
| Specific radioactivity (counts/mg protein) | | | | |
| "Flight," 2d day | 48±2 | 38±1* | 47±3* | 58±3* |
| Control | 49±2 | 47±2 | 59±2 | 89±1 |
| "Synchronous," 2d day | 76±6 | 71±4 | 53±5 | 44±2* |
| Control | 64±2 | 64±4 | 47±2 | 56±2 |
| "Flight," 26th day | 44±4 | 41±3 | 33±2 | 54±4 |
| Control | 43±2 | 37±2 | 33±1 | 45±2 |
| "Synchronous," 26th day | 50±2 | 38±2 | 49±2 | 48±4 |
| Control | 52±2 | 40±1 | 48±1 | 54±2 |
| Total radioactivity (counts/min/muscle) | | | | |
| "Flight," 2d day | 192±27 | 185±22* | 168±16* | 243±35 |
| Control | 256±21 | 304±36 | 286±23 | 404±71 |
| "Synchronous," 2d dat | 356±46 | 326±27 | 289±22 | 240±32 |
| Control | 340±9 | 390±18 | 287±15 | 314±23 |

Note: The means of 4-5 readings on experimental animals and 6-7 readings on vivarium control rats are listed.

Thus, we succeeded in demonstrating changes that are typical of the effects of weightlessness in both red (tonic) and white (phasic) skeletal muscles. Atrophy of red muscle, demonstrable only in "flight" rats, is apparently due to faster breakdown of proteins, since we failed to demonstrate inhibition of biosynthesis of protein molecules in this muscle, at any rate not at the times we examined it. Although a moderate atrophic reaction is observed in both "flight" and "synchronous" animals, in white muscle, at the early stages after termination of exposure to the specific experimental conditions involved, the mechanisms upon which this reaction is based are apparently not the same. While synthesis of protein molecules is depressed in this muscle under the influence of weightlessness, this does not occur under the effect of the conditions of the synchronous experiment. As a result, both faster breakdown and inhibition of protein synthesis may

play a role in the pathogenesis of atrophy of white muscle in the former instance, this applies primarily to intensified degradation of protein molecules in the latter.

Typically enough, the reaction of the protein-synthesizing system of the muscles we examined to weightlessness or hypokinesia, in the synchronous experiment, differs from the reaction observed in the case of "disuse" of the same muscles under model conditions, with exarticulation of the foot and immobilization of distal tendons in the region of the epiphyses of the tibia and fibula. In this case, marked atrophy of red muscle is associated with severe inhibition of protein synthesis in it, while moderate atrophy of white muscle is related to appreciable activation of this process [4].

The submitted data warrant the conclusion that a reversible, differentiated reaction is observed in muscles of different functional types to the set of factors of space flights and the synchronous experiment. This differentiation is manifested by both the difference in their susceptibility to atrophy and the distinctive metabolic mechanisms of development thereof.

BIBLIOGRAPHY

1. Portugalov, V. V. in: "Tezisy dokladov simpoziuma 3-go Vsesoyuznogo biokhimicheskogo s"yezda" [Summaries of Symposium Papers Delivered at the 3d All-Union Biochemical Congress], Riga, 1974, pp 317-318.

2. Ivanov, I. I., and Yur'yev, V. A. "Biochemistry and Pathological Biochemistry of Muscles," Leningrad , 1961, p 241.

3. Lowry, O. M.; Rosebrough, N. I.; Farr, A. L.; et al. J. BIOL. CHEM., Vol 193, 1951, pp 265-267.

4. Kazar'yan, V. A.; Goncharova, L. A.; and Rapoport, E. A. PAT. FIZIOL. [Pathological Physiology], No 4, 1974, pp 45-49.

UDC: 612.014.461.3+612.015.31]-06:629.78

# FLUID-ELECTROLYTE METABOLISM IN ALBINO RATS AFTER A FLIGHT ON THE KOSMOS-690 BIOSATELLITE

Moscow KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA in Russian
No 6, 1977 pp 23-26

[Article by N. A. Ilyushko, Ye. A. Il'in, Yu. I. Kondrat'yev, V. I. Korol'kov, L. V. Kokoreva and Yu. N. Khodkevich, submitted 13 Jul 76]

[Text] Most researchers believed that the changes in fluid-electrolyte metabolism observed in crew members of spacecraft are due to the effects of weightlessness [1-4].

In this work, we studied the indices of fluid-electrolyte metabolism in albino rats after an orbital flight on the Kosmos-690 biosatellite, with exposure to space flight factors. The fact that maintenance conditions, duration of flight and diet were identical on Kosmos-605 and Kosmos-690 made it possible to make a comparative analysis of the changes in some indices of fluid and electrolyte metabolism in the readaptation period [5].

## Methods

The animals received homogeneous, biologically complete feed. The constant chemical composition of the feed, its homogeneous, pasty consistency, which ruled out the possibility of loss when feeding the animals, made it possible to conduct balance studies in the readaptation period. For this purpose, metabolic studies were made, with precise records of feed and water intake, as well as output of feces and urine, on the 2d-3d, 4th-5th, 11th-12th and 20th-21st days after the flight on Kosmos-690 in constant groups of 5-7 animals exposed to 800 rad radiation. The same protocol was followed in the experiment on the Kosmos-605 satellite. Determination was made of water intake, intake of fluid in feed, urine excretion, balance of potassium, sodium and calcium. In order to evaluate the overall hydration level, we calculated the hydration coefficient, which is the ratio of all fluid intake to excreted urine per 100 g body weight. We used the method of flame photometry to assay potassium, sodium and calcium in feed and biological specimens.

Results and Discussion

According to the data in Table 1, there was an increased intake of drinking water and reliable elevation of hydration coefficient in flight animals and rats in the "control-2" experiment up to the 12th day, as compared to intact animals, which may be indicative of water retention in this period. We failed to demonstrate significant differences in urine excretion in all groups and at all examination times.

Table 2 submits data on sodium balance in the readaptation period. Retention of sodium in "flight" animals on the 2d-3d day constituted 21% of all sodium taken with feed and on the 11th-12th day, 29%. Virtually the same changes were noted in sodium balance in "control-2." At the same times, sodium retention constituted 24 and 30%, respectively, in these animal groups. The studies of sodium balance in "flight" and "control-2" animals warranted the assumption that the high retention of sodium in the readaptation period may be largely attributable to loss thereof in the course of the flight and model experiments. The absence of reliable differences in sodium balance indicates that the conditions under which the animals were kept, rather than weightlessness, were the cause of sodium deficiency in the flight and ground-based "control-2" experiment. It must be mentioned that the minor sodium retention after the Kosmos-609 flight is not observed from the very first day of readaptation, as is the case after the Kosmos-605 flight; it was demonstrated only on the 11th-12th day, which is indicative of more significant impairment of regulatory and adaptation mechanisms of the animal organism under the combined influence of maintenance conditions and exposure to 800 rad radiation.

The combination of space flight factors and irradiation had a substantial effect as well on potassium metabolism of "flight" animals. Thus, after the Kosmos-690 flight, the rats presented a negative potassium balance up to the 5th readaptation day. After the Kosmos-605 flight, a negative potassium balance was observed only on the 2d day of readaptation, and already on the 3d-5th and 11th-12th days we observed maximum retention of potassium. Thus, while restoration of potassium level began on the 3d readaptation day after the Kosmos-605 flight, maximum potassium retention was noted only by the 11th-12th day after the Kosmos-690 flight.

In all of the groups, the calcium balance was positive after the flight on the Kosmos-690 biosatellite. However, it should be noted that there was maximum calcium retention on the 2d-5th and 11th-12th days of the readaptation period. It constituted 29% on the 2d-5th day and 34% of all calcium that was taken in on the 11th-12th day in the "flight" rats. Calcium retention was the same on the 2d-5th and 11th-12th days, constituting 31%, in "control-2," and the figures were 21 and 23%, respectively, in the intact control. Elimination of calcium on the 2d-5th day of readaptation was at a lower level in flight animals ($P<0.05$) and rats in the "control-2" experiment ($P<0.01$), as compared to intact rats.

Table 1. Fluid intake and output in readaptation period after Kosmos-690 flight (M±m)

| Day of examination | Animal group | Water intake, mℓ | Total fluid mℓ | Fluid excreted in urine, mℓ | Hydration coefficient | P |
|---|---|---|---|---|---|---|
| 2—3 | "flight" | 17,1±2,78 | 42,5±3,17 | 25,1±4,47 | 0,70±0,04 | $P_2<0,01$ |
| | "control-2" | 18,6±3,77 | 42,9±4,12 | 22,9±4,38 | 0,70±0,09 | $P_3<0,01$ |
| | intact control | 8,9±3,79 | 36,6±3,79 | 25,1±3,79 | 0,50±0,04 | |
| 4—5 | "flight" | 20,4±4,07 | 45,4±4,30 | 30,6±4,50 | 0,60±0,03 | $P_2<0,01$ |
| | "control-2" | 18,1±3,55 | 44,3±3,83 | 27,1±4,60 | 0,60±0,03 | $P_3<0,01$ |
| | intact control | 12,7±4,23 | 40,4±4,23 | 29,6±4,41 | 0,45±0,03 | |
| 11—12 | "flight" | 21,4±2,79 | 49,4±3,66 | 23,1±0,96 | 0,79±0,09 | $P_2<0,01$ |
| | "control-2" | 20,4±4,00 | 46,5±3,55 | 27,6±3,85 | 0,60±0,04 | $P_3<0,01$ |
| | intact control | 12,6±4,14 | 41,0±4,14 | 28,4±5,11 | 0,47±0,04 | $P_1<0,01$ |
| 20—21 | "flight" | 27,5±2,09 | 54,9±3,35 | 34,2±4,02 | 0,55±0,06 | |
| | "control-2" | 20,1±4,17 | 46,8±4,04 | 32,2±4,62 | 0,46±0,04 | |
| | intact control | 13,4±3,20 | 41,8±3,20 | 26,8±2,46 | 0,46±0,01 | |

$P_1$) "flight" group compared to "control-2"
$P_2$) same compared to intact control
$P_3$) "control-2" compared to intact control



Table 2. Sodium and Potassium balance in readaptation period after Kosmos-690 and Kosmos-605 flights (M±m)

| Day of examination | Animal group | Kosmos-690 sodium | | Kosmos-605 sodium | | Kosmos-690 potassium, | Kosmos-605 potassium, |
|---|---|---|---|---|---|---|---|
| | | mg | % | mg | % | mg | mg |
| 2—3 | "flight" | +17,8±5,42 | 21 | +25,1±4,91 | 33 | —1,7±3,75 | —3,3±2,34 |
| | "control-2" | +16,0±3,41 | 24 | +24,9±3,01 | 30 | +5,6±2,48 | +5,8±1,65 |
| | intact control | +14,5±3,29 | 18 | +15,4±2,57 | 18 | +5,0±1,48 | +6,2±1,28 |
| 4—5 | "flight" | +16,9±4,22 | 20 | +37,8±4,25 | 47 | —0,36±0,45 | +22,0±2,62 |
| | "control-2" | +17,5±2,84 | 23 | | | +4,2±2,04 | |
| | intact control | +15,1±1,56 | 17 | +15,6±2,83 | 20 | +4,1±1,44 | +5,5±0,96 |
| 11—12 | "flight" | +25,5±1,80 | 29 | +19,5±3,65 | 24 | +10,1±2,72 | +7,2±1,53 |
| | "control-2" | +27,8±1,25 | 30 | | | +6,2±1,23 | |
| | intact control | +13,7±6,35 | 16 | +15,4±1,79 | 19 | +4,6±1,62 | +3,3±0,83 |
| 20—21 | "flight" | +12,6±2,12 | 15 | +12,2±2,16 | 16 | +5,4±1,98 | +5,8±1,60 |
| | "control-2" | +13,5±1,31 | 16 | +13,0±1,34 | 15 | +4,0±1,98 | +4,5±0,96 |
| | intact control | +13,0±2,66 | 15 | | | +5,1±1,12 | +3,7±0,97 |

The data obtained on fluid and electrolyte retention in the postflight period can be evaluated as a compensatory reaction of the organism, related to making up the deficiency that developed during the flight. Evidently the loss of potassium in the first postflight stage can be interpreted as a continuation of loss of potassium ions under the influence of weightlessness. Elimination of potassium from the organism in weightlessness is related primarily to loss of intracellular potassium [1].

Most of the changes described were reversible. Data close to the ones referable to intact animals were obtained on the 20th-21st day of the readaptation period.

Thus, the study of the nature of metabolic processes referable to fluid and electrolytes in the postflight period, in rats, enabled us to establish several features in common in reactions of animals and man to space flight factors. At the same time, the longer period of recovery of the parameters studied (hydration of the organism, balance of potassium, sodium and calcium) after the Kosmos-690 flight may be indicative of diminished reactivity of the organism due to the combined effect of weightlessness and 800 rad of radiation.

BIBLIOGRAPHY

1. Balakhovskiy, I. S., and Natochin, Yu. V. in: "Problemy kosmicheskoy biologii" [Problems of Space Biology], Moscow, Vol 22, 1973, pp 1-116.

2. Leach, C. S. in: "Skylab Preliminary Biomedical Report," (NASA), Houston, Sect 3, 1973, pp 177-190.

3. Lutwak, L.; Whedon, G. D.; et al. J. CLIN. ENDOCR., Vol 28, 1969, p 1140.

4. Whedon, G. D.; Lutwak, L.; Ranbauf, P. C.; et al. in: "Skylab Preliminary Biomedical Report," (NASA), Houston, Sect 3, 1973, pp 169-176.

5. Ilyushko, N. A.; Il'in, Ye. A.; Korol'kov, V. I.; et al. in: "Aviakosmicheskaya meditsina. Tez. dokl. 5-y Vsesoyuznoy konferentsii" [Aerospace Medicine. Summaries of Papers Delivered at the 5th All-Union Conference], Moscow--Kaluga, Vol 1, 1975, pp 12-14.

UDC: 616.8-02:612.766.2

# CLINICAL ASPECTS OF CHANGES IN THE NERVOUS SYSTEM IN THE COURSE OF 49-DAY ANTIORTHOSTATIC HYPOKINESIA

Moscow KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA in Russian
No 6, 1977 pp 26-31

[Article by T. N. Krupina and A. Ya. Tizul, submitted 30 May 75]

[Text] Specific hemodynamic changes occur during space flights, during the period of cosmonaut adaptation to weightlessness [1-6]. Authors who are concerned with the effects of hypokinesia [7-28] believe that the hemodynamic changes are similar to those observed in cosmonauts during weightlessness. More recently, the model of clinostatic hypokinesia has been upgraded with a new component, antiorthostatic position with the head of the bed tilted down at an angle of 4-6° [29-31].

We studied the condition of the nervous system of 9 individuals ranging in age from 20 to 35 years in the course of 49-day antiorthostatic hypokinesia. In addition to clinical tests, we studied thermoregulation and the muscular system once every 7-10 days. We questioned the subjects about their well-being daily.

Analysis of the clinical and special studies revealed that the common finding made in all subjects was that they felt heat and a rush of blood to the head within 15-20 min from the start of the study, and this reached a maximum after 1.5 h. Most subjects developed other symptoms as well 4-5 h later: congested nose and ears, heaviness of the head and a sensation of pressure on the eyeball. The sensations due to downward tilting of the head of the bed were evaluated in different ways: ranging from the sensation of mild rush of blood to the head at the very start to compression and distensive pain during the period of maximal expression thereof. The voice dropped to a lower pitch in some subjects. Difficulty in breathing and a sensation of retrosternal compression were observed in two subjects. Most of the subjects had no trouble falling asleep. On the 2d day, there was less sensation of blood rushing to the head; a mild heaviness of the head and congested nose and ears remained.

It should be noted that for the first 1-2 days of hypokinease there was increased diuresis and vesical tenesmus in all of the subjects.

By the end of the 2d day, most subjects developed signs of physical discomfort (pain in the back, lumbar region, uncomfortable lying down, cold feed, restlessness). For 2-4 days the signs of physical discomfort progressed, then began to regress and disappeared entirely by the 6th-8th day; the subjects appeared to get used to the conditions. At the same time, they had lost, almost completely, the "angular feeling" of the head-down position. The prominent complaint at this time was referable to temperature, as manifested by cold sensation in the distal parts of the limbs, particularly the fee , which was more marked in the daytime (starting at 1000-1100 hours).

Objectively, no changes were noted in the neurological status for the first 3 weeks of the study. In the 4th week, which is somewhat sooner than in the case of clinostatic hypokinesia [11], some subjects presented solitary nystagmoid beats with the eyeballs in extreme positions. By the end of the 5th week, nystagmus was observed in virtually all of the subjects. In addition, there was activation of tendon and periostal reflexes, not infrequently with signs of asymmetry (more accentuated on the right in 5 subjects and on the left in 2 others, there being no clearcut difference between the two sides in the other 2 subjects), mild digital tremor while performing locomotor acts, change in nature and duration of autonomic reflexes (dermographism, Aschner reflex, Laignel-Lavastine syndrome). These neurological changes were observed in all subjects to varying extents, and they presented a tendency toward progression up to the 40th-44th day of the study. Most subjects became irritable and were in a bad mood starting in the 5th week of hypokinesia; some of them stated that "I'm tired of lying down," "I do not feel like doing anything," and "I've lost interest in the study"; they were no longer interested in books, they were unwilling to come in contact with others, and most often they responded formally and briefly to questions about their wellbeing. After the 30th day, some stated: "more and more often I realize that I have to use self-control not to break down," "I wish this would end soon." Their sleep worsened. They began to be irritated by tests and trifles that they did not notice previously. This subjective signs of asthenia progressed up to the 45th-46th day of the study, after which there was some emotional elation, apparently due to the fact that the end of the study was near.

Objectively, during the period of asthenization, there were some changes in the nervous system, particularly the autonomic status. Thus, there was a mean increase in pulse rate of 15/min (Table 1). There was no change in mean systolic pressure. Diastolic pressure rose by 10-20 mm Hg in 7 out of 9 subjects and dropped by 10-15 mm Hg in the 2 others. A pressure drop was observed only in the subjects who presented somewhat high diastolic pressure in the background period (85-95 mm Hg).

There was a change in nature and duration of dermographic reactions. While there was prevalence of the red phase of local dermographis in the background period and at the start of the study, this applied to the white phase during the period of hypokinesia. During the first few days of the recovery period there was prevalence of the white phase, like in the hypokinetic period;

thereafter, both the white and red phases of dermographic reactions were observed. The duration of the "white spot" phenomenon increased in the course of the study and reached 6 s toward the end of the hypokinetic period.

Table 1. Dynamics of changes in some autonomic indices during the experimental and recovery periods (mean data)

| Index | | Background | Day of hypokinesia | | | | | Difference | Day of recovery period | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | 7 | 15 | 25 | 35 | 49 | | 3 | 13 | 29 |
| Pulse | | 67 | 72 | 77 | 75 | 80 | 82 | +15 | 77 | 92 | 71 |
| Arterial pressure | systolic | 126 | 123 | 123 | 123 | 122 | 124 | —4 | 125 | 123 | 122 |
| | diastolic | 75 | 77 | 81 | 84 | 82 | 85 | +10 | 83 | 82 | 81 |
| Duration of dermographism, s | white phase | — | — | 95,7 | 74,4 | 90 | 92,7 | — | 84,4 | 54,1 | 66,6 |
| | red phase | 89 | 72,3 | — | — | — | — | — | — | 72,2 | 62,8 |
| Duration of "white spot" phenomenon, s | right | 3,2 | 3,4 | 3,8 | 4,1 | 4,2 | 4,2 | +2 | 4,0 | 4,0 | 3,6 |
| | left | 3,2 | 3,4 | 3,9 | 3,9 | 4,0 | 4,1 | +0,9 | 4,1 | 4,0 | 3,8 |

In the course of the study, we observed some correlation between changes in vegetative indices. Thus, lability of pulse with a tendency toward acceleration and elevation of diastolic pressure coincided with prevalence of the white phase of dermographism and longer duration of the "white spot" phenomenon. These clinical symptoms could be indicative of some predominance of sympathetic tonus during the hypokinetic period and development of unstable autonomic regulation, particularly since we also observed a 3-7°C temperature drop in the distal parts of the limbs, as compared to background levels; with the change in nature and duration of dermographism there appeared complaints of chilliness and cold in the distal parts of the limbs.

The study of thermoregulatory function, based on daily thermometry of the axillary region and analysis of indices of Shcherbak's thermoregulatory reflex revealed a temperature drop (by a mean of 0.35°C) in the axillary region, as well as change in thermoregulatory dynamics, as manifested by sluggishness or areactivity of thermoregulatory processes with the use of a local thermal load according to Gauffe. These data are apparently indicative of functional changes in central (hypothalamic) elements of regulation, which are responsible for thermoregulatory dynamics.

Clinical observations also revealed that with increase in duration of both clinostatic and antiorthostatic hypokinesia there is development of trophic

Table 2. Dynamics of recovery of force indices of various muscle groups according to polydynamometry findings (mean data)

| Extensors | | | | | Flexors | | | |
|---|---|---|---|---|---|---|---|---|
| muscle group | index | background | recovery day | | muscle group | background | recovery day | |
| | | | 4 | 10 | | | 4 | 10 |
| Forearm | $M \pm m$ | 24,4±1,9 | 22,4±1,5 | 24,7±1,2 | Forearm | 35,5±2,4 | 34,8±1,7 | 37,8±1,9 |
| | $\sigma$ | 5,5 | 4,3 | 3,3 | | 6,7 | 4,7 | 5,4 |
| Arm | $M \pm m$ | 63,3±3,6 | 62,2±4,8 | 61,3±1,7 | Arm | 37,7±3,1 | 35,0±1,9 | 38,2±1,8 |
| | $\sigma$ | 10,4 | 13,5 | 5,0 | | 8,7 | 5,4 | 5,1 |
| Thigh | $M \pm m$ | 127,4±12,3 | 110,2±6,6 | 125,1±4,7 | Thigh | 27,5±1,9 | 26,5±0,9 | 28,1±1,3 |
| | $\sigma$ | 34,7 | 18,8 | 13,5 | | 5,4 | 2,7 | 3,7 |
| Leg | $M \pm m$ | 42,5±3,1 | 38,8±1,8 | 34,1±2,1 | Leg | 11,8±1,8 | 10,1±1,4 | 10,5±1,1 |
| | $\sigma$ | 8,7 | 5,0 | 6,0 | | 5,04 | 4,04 | 3,03 |
| Trunk | $M \pm m$ | 151,4±5,7 | 118,7±8,4 | 128,7±19,8 | Trunk | 129,3±7,9 | 102,4±6,3 | 129,4±4,0 |
| | | 16,2 | 23,9 | 14,1 | | 22,5 | 17,8 | 11,4 |

disturbances, such as dryness of the integument, particularly of the legs, brittle nails, subatrophic changes in the mucosa of the upper respiratory tract (in spite of plethora thereof), as well as hypotrophy (weight loss) of muscles, more marked in the lower limbs (the diameter of the legs decreased by 4 cm).

The tests of muscle tonus according to Sarmai revealed that as time passed the tonus of all muscles diminished; this was the most significant in the distal parts of the lower extremities.

The muscular strength of the right hand diminished by a mean of 12.9 kg by the end of the hypokinetic period. The most severe decrease was observed in repeated tests with the squeeze dynamometer. This suggested that, along with decline of force indices, there was the phenomenon of muscular fatigue.

We also observed a decline of force indices according to polydynamometry data in most of the muscle groups examined (Table 2).

There was restoration of force indices to base levels by the end of the first 10 days of the recovery period in the muscle groups that do not carry a postural tonic load. The force indices were not restored by the 10th day in muscle groups carrying the largest load with regard to implementation of gravitational tonic-postural reflexes (muscles of the trunk, legs and feet).

We observed a very severe pain syndrome, mainly in the muscles of the lower limbs and foot joints, from the first days of the recovery period, related to the change from horizontal to vertical position and start of exercise. In addition, all of the subjects suffered from vertigo and intensification of neurological

symptoms when they stood up (particularly on the first 1-2 days): impaired gait, instability in Romberg's station, impaired regulation of vertical position, palpebral and digital tremor, more accentuated asymmetry of tendon reflexes (D>S in 5 cases, S>D in 2, and no clearcut difference in 2 cases) and diminished abdominal reflexes. All of the subjects presents signs of vegetovascular dysfunction and asthenization of the organism, manifested by general weakness, fatigability, faster pulse, to 120-130/min, severe change in heart rate and arterial pressure with change in body position (clinostatic and orthostatic reflexes), diminished orthostatic stability, change or distortion of Aschner-Dagnini and Chermak reflexes, regional or generalized hyperhidrosis.

Thus, on the basis of analysis of the dynamics of adaptation processes associated with antiorthostatic hypokinesia, we can arbitrarily distinguish three phases: the so-called "acute" phase, related to reorganization of the circulatory system in response to acute redistribution of blood, a phase of relative "adaptation" and readaptation phase.

The clinical symptoms that develop in the "acute" phase can be attributed to redistribution of blood and reorganization of the circulatory system. This has been demonstrated by Kh. Kh. Yarullin et al. [31], who made a rheographic study of regional circulation. Thus, in the "acute" phase (1st week), there is diminished tonus of vessels of the head, bones and legs, with signs of overload on the pulmonary circulatory system and venous stasis in the head. Thereafter, as a result of stimulation of baroreceptors related to venous stasis, the phase of relative adaptation begins in the hemodynamic system. The diminished load and tone of skeletal muscles [9, 11, 12, 14, 16-28] is another substantial pathogenetic factor that elicits changes in the organism; this, in turn, leads to a change in functional state of the nervous system and mediated changes in regulation of metabolic processes and functions of the hypothalamo-hypophyseo-adrenal system.

All of the foregoing determines the complex and variable pattern and polymorphism of changes in various systems of the organism, which are observed under the influence of factors involved in antiorthostatic hypokinesia. Among the changes in the nervous system, the following syndromes are the ones that are observed the most consistently with antiorthostatic hypokinesia: vegetovascular dysfunction, asthenoneurotic syndrome and syndrome of neuromuscular disturbances. The "acute" phase is more marked with antiorthostatic hypokinesia, as opposed to clinostatic, while there is somewhat earlier development of changes referable to the nervous system in the phase of relative "adaptation." In the "acute" stage of readaptation, statokinetic disturbances and a very marked pain syndrome in the joints and muscles of the lower limbs and back are added to these syndromes. The longest time is required for recovery from the asthenoneurotic syndrome and signs of vegetovascular instability.

BIBLIOGRAPHY


1. Vorob'yev, Ye. I.; Nefedov, Yu. G.; Kakurin, L. I.; et al. KOSMICHESKAYA BIOL. [Space Biology], No 4, 1969, pp 46-50.

2. Idem, Ibid, No 2, 1970, pp 65-74.

3. Molchanov, N. S.; Krupina, T. N.; et al. Ibid, No 6, 1970, pp 39-42.

4. Kovalenko, Ye. A., and Vasil'yev, P. V. IZV. AN SSSR. SERIYA BIOL. [News of the USSR Academy of Sciences. Biology Series], No 3, 1971, pp 356-369.

5. Berry, Ch. AEROSPACE MED., Vol 40, 1969, p 245.

6. Gazenko, O. G. in: "Problemy kosmicheskoy biologii" [Problems of Space Biology], Moscow, Vol 13, 1969, pp 5-7.

7. Kakurin, L. I. KOSMICHESKAYA BIOL., No 2, 1968, pp 59-63.

8. Genin, A. M.; Sorokin, P. A.; et al. in: "Problemy kosmicheskoy biologii," Moscow, Vol 13, 1969, pp 247-253.

9. Kalin, G. S., and Terent'yev, V. G. Ibid, pp 214-220.

10. Krupina, T. N.; Tizul, A. Ya.; et al. KOSMICHESKAYA BIOL., No 5, 1967, pp 61-66.

11. Krupina, T. N., and Tizul, A. Ya. ZH. NEVROPATOL. I PSIKHIATR. [Journal of Neuropathology and Psychiatry], No 11, 1971, pp 1611-1617.

12. Panov, A. G.; Lobzin, V. S.; and Mikhaylenko, A. A. Ibid, No 4, 1973, p 506.

13. Parin, V. V.; Krupina, T. N.; Mikhaylovskiy, G. P.; et al. KOSMICHESKAYA BIOL., No 5, 1970, pp 59-65.

14. Tizul, A. Ya.; Sokolov, Ye. I.; Mdinaradze, Yu. S.; et al. SOV. MED. [Soviet Medicine], No 7, 1972, pp 147-148.

15. Tizul, A. Ya.; Kozlov, B. V.; and Anan'yev, G. V. KOSMICHESKAYA BIOL., No 4, 1972, pp 72-77.

16. Kakurin, L. I. "Effects of Restricted Exercise on the Human Body," author abstract of doctoral dissertation, Moscow, 1968.

17. Portugalov, V. V.; Gazenko, O. G.; et al. KOSMICHESKAYA BIOL., No 6, 1967, pp 18-25.

18. Sorokin, P. A.; Simonenko, V. V.; and Korolev, B. A. "Problemy kosmicheskoy biologii," Moscow, Vol 13, 1969, p 24.

19. Chase, G.; Grave, C.; and Rovel, R. AEROSPACE MED., Vol 37, 1966, p 1232.

20. Graveline, D., and Bernard, G. Ibid, Vol 32, 1961, p 726.

21. Graveline, D., and McCally, M. J. APPL. PHYSIOL., Vol 17, 1962, p 519.

22. Kraus, H., and Raab, W. "Hypokinetic Disease," Springfield, 1961.

23. Lamb, L. AEROSPACE MED., Vol 35, 1964, p 413.

24. Lamb, L., and Stevens, P. Ibid, Vol 36, 1965, p 1145.

25. Miller, P.; Hartman, B.; et al. Ibid, Vol 35, 1964, p 1194.

26. Miller, P.; Johnson, R.; et al. Ibid, p 391.

27. Miller, P.; Johnson, R.; and Lamb, L. Ibid, Vol 36, 1965, p 1077.

28. Vogt, F.; Mack, P.; Johnson,R.; et al. Ibid, Vol 38, 1967, p 43.

29. Voskresenskiy, A. D.; Yegorov, B. B.; et al. KOSMICHESKAYA BIOL., No 4, 1972, pp 28-32.

30. Genin, A. M., and Kakurin, L. I. Ibid, No 4, 1972, pp 26-28.

31. Yarullin, Kh. Kh.; Krupina, T. N.; Vasil'yeva, T. D.; et al. Ibid, pp 33-39.

UDC: 612.748-06:612.766.2

# STATE OF RAT MUSCLE MOTONEURON SYSTEM IN THE CASE OF RESTRICTED MOBILITY

Moscow KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA in Russian
No 6, 1977 pp 31-35

[Article by Ye. I. Il'ina-Kakuyeva and V. V. Portugalov, submitted 4 Sep 75]

[Text] It is a known fact that restricted motor activity has an adverse effect on the human body. This condition is becoming an increasing part of everyday life of healthy individuals; it is also present in a sick individual and sometimes aggravates the course of the chief disease. Cosmonauts involved in space flights are still susceptible to it. Studies on man and animals have resolved a number of cardinal problems related to hypokinesia; however, there are still numerous special questions to which answers must be given.

Our previous studies [1-3] revealed that hypokinesia in rats leads to significant structural and metabolic changes, selectively in the soleus muscle, the muscle fibers of which are red. The pathological process that develops in this muscle presents distinct dynamics. The stages of development thereof are related to long-term hypokinesia. At the early stages (1st-7th days) of hypokinesia, edema and proliferation of connective tissue elements develop in the muscular endomysium, the muscle fibers swell and some of them are subject to destructive changes leading to death thereof. The process is usually focal, but it can extend over the entire muscle. The next phases of development of the process, which successively follow one another, are characterized by formation of so-called target fibers (7th-30th days) and transformation of the latter into "target-like fibers" (45th-60th days), so named because of the unique distribution of the product of histochemical reaction, which is formed upon demonstration of oxidative enzymes, not uniformly in the sarcoplasm of the fiber, which is the usual finding, but in its medial or peripheral region [4, 5]. The last stage of the process takes place against the background of marked atrophy. Since "target fibers" are observed in muscles in the presence of neurogenic myopathy [5, 6], the question arises as to whether disturbances referable to innervation are the basis of such changes. The information in the literature concerning the possible cause of this form of muscular pathology is rather hypothetical in nature [5, 6].

Appearance of target fibers is not observed in muscle fibers of the gastrocnemius muscle, which is functionally similar to the soleus but differs in chemistry. In the former, primarily atrophic processes develop during hypokinesia, rather than dystrophic. In this regard, another question arises: What is the state of the motor nerve elements of these muscles, which react so differently to restricted mobility, in the presence of hypokinesia?

Methods

Experiments were conducted on white, mongrel, male rats kept in small individual cages for 7, 15, 30, 45 and 65 days. We used 10 animals for each testing time. The same number of rats served as a control for each such time. The nerve structures of the soleus and gastrocnemius were demonstrated by the method of Bielschowsky-Gross in the modification of Campos. In addition, muscle sections were studied by histological techniques. We demonstrated succinate dehydrogenases and $NADH_2$ in cryostat sections of nonfixed specimens of muscles for identification of "target fibers" and "target-like fibers."

Results and Discussion

Examination of muscle sections impregnated with silver salts revealed that changes occur in motor nerve endings of muscle tissue. On the 7th-15th days of hypokinesia, when "target fibers" begin to develop and some muscle fibers perish, there are considerable changes in motor nerve endings, to the extent of total degeneration. On the 7th day of hypokinesia, some end plates present destruction of sole plates and degeneration of terminal elements of nerve fibers. The sole plate cells acquire a vague outline and later change into a homogeneously stained mass. In such plates, the nerve endings become coarser, thicker and separate into argentophilic clumps (see Figure, f, g). The cells of sole plates, situated in the region of dead muscle fibers, among proliferative connective tissue elements, have the appearance of homogeneously stained dense masses, near which there are argentophilic clumps of fragmented nerve fiber endings.

On the 30th experimental day, after formation of "target fibers" is completed, the changes in motor endings vary in nature. Myoneural endings are encountered, the sole-plate cells of which consist of a homogeneously stained mass, while the terminal branches of nerve fibers appear as thickened stumps. Terminal segments of nerve fibers of this kind are demonstrable on muscle fibers in a state of degeneration. Motor plates, the nervous system of which is represented by profusely arborized coarse terminals, are seen on muscle fibers that appear unchanged on silver-impregnated preparations; in such plates, the outlines of sole cells are vague (see Figure, h). A typical finding is the presence of nerve endings that go beyond the borders of the motor plates and extend along the muscle fibers, giving off free lateral branches along their course (see Figure, i). Myelinated fibers are encountered, which give off thin processes in the preterminal segments, terminating here on a muscle fiber or passing beyond it. The absence of connections with ancillary cells is inherent in virtually all altered motor endings. As a rule, such findings are not made in the case of briefer hypokinesia.

Since altered motor plates are encountered only among injured muscle fibers, the number of such plates is directly related to the amount of muscle tissue involved in the pathological process.

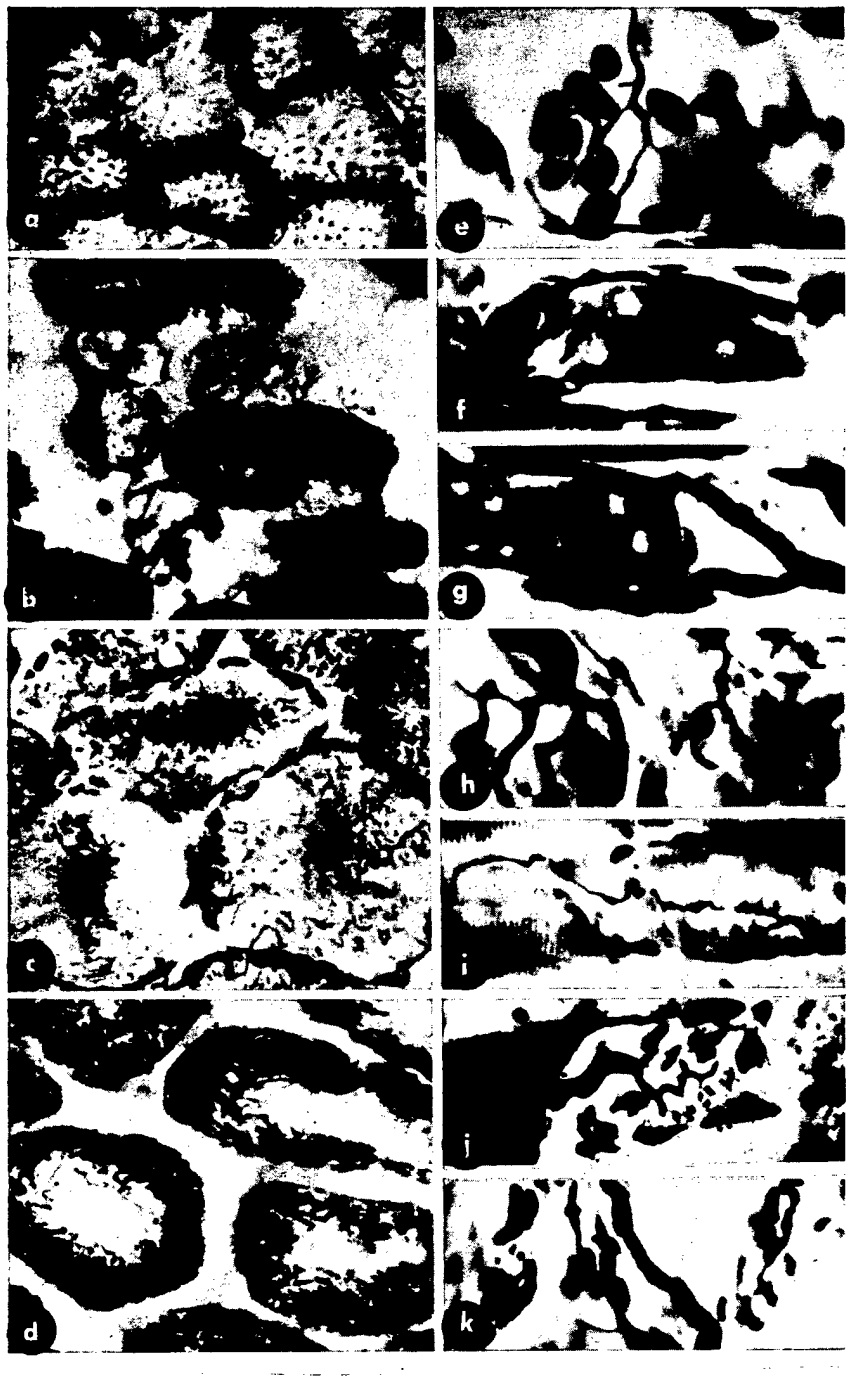
On the 45th-65th days of hypokinesia, when the process of formation of "target-like fibers" takes place, we still encounter myoneural elements characterized by excessive arborization of nerve endings, "coarsening" thereof or with thin processes taking off from the preterminal segment, which do not come in contact with the sole plate cells (see Figure, j); however, there are considerably fewer such processes than on the 30th day of hypokinesia. On the 65th day of the experiment, there is prevalence of neuromuscular endings in the muscle that do not differ from the motor plates of control animals, as well as small plates that are not demonstrable at other times (see Figure, k).

Special mention should be made of the fact that, at all stages of hypokinesia, the changes were referable only to terminal and preterminal segments of motor fibers.

Unlike the soleus muscle, the gastrocnemius failed to demonstrate any appreciable changes in myoneural elements.

When the rats are kept in cages that restrict their mobility, structural changes develop in the soleus, which extend not only to muscle tissue, but to the peripheral motor nerve system that implements its function. These facts evidently indicate that the changes in the soleus muscle, which lead to formation of "target fibers," are very closely linked with structural disturbances in the motor endings that innervate them. At the same time, we have no grounds for making a categoric statement as to the nature of the mechanisms upon which the above-described changes in muscles are based. Since the motor plates subject to degeneration are encountered primarily in sites of destructively altered muscle tissue, the question arises as to whether metabolic and structural disturbances occur first in muscle fibers proper, for example, under the influence of ischemia that develops for some reason. We have reason to believe that development of focal edema in the soleus muscle on the 1st day of hypokinesia is the triggering mechanism that leads to development of metabolic and structural changes in all components of muscle tissue and destruction of some muscle fibers. As a result, the motor nerve endings lose their contact with dying muscle fibers. The latter, in turn, are no longer under the trophic influence of the nervous system, and this aggravates the severity of the process taking place in them.

The stage of predominance of destructive processes in the muscle is followed, on the 30th day of the experiment, by a different stage, which is characterized by signs indicative of repair processes, that justify our reference to reinnervation of muscle fibers. This is indicated by signs of "excessive growth" in the region of the endings of motor nerve conductors, formation of atypical terminal nerve elements, appearance of processes branching away from the preterminal segments of neural conductors that have no structural disturbances.



Soleus muscle. Distribution of NAD-$H_2$ dehydrogenase activity

a) in control animal
b) degenerative fibers with 7-day hypokinesia [HK]
c) "target fibers" with 30-day HK
d) "target-like fibers," 65-day HK

Motor nerve endings:

e) of control animal
f, g) with 7-day HK
h, i) 30-day HK
j, k) 65-day HK

Magnification:

a-b) objective, 20×; ocular, 6.3×
c-k) objective, 40×; ocular, 6.3×

Silver salts impregnation according to Bielschowsky-Gross as modified by Campos.

Processes of both a destructive and reparative nature may be demonstrated in the soleus muscle by the 65th day of experimental restriction of movement; and, according to the condition of the terminal elements, there are more extensive signs of the latter process. The period of development of repair processes coincides with the time of formation of "target-like fibers" from the "target fibers," i.e., apparently with the period of muscular adaptation to limited mobility.

In the presence of hypokinesia, the changes in the motor plates are not specific, and they are encountered in the presence of destructive and repair processes induced by other causes [7, 8, 9].

Neither "target fibers" nor structural disturbances of motor nerve elements are demonstrable in the gastrocnemius during hypokinesia. In the case of restricted mobility, the changes in this muscle are analogous to those observed in muscle tissue in the presence of simple atrophy. It is difficult to state at this time what causes different changes in muscular structures and nerve elements in these two muscles that are similar in function. Perhaps the difference is related to the distinctions referable to blood delivery to the two muscles. Edema develops in the soleus, which has a more developed capillary network than the gastrocnemius. Perhaps, also, the differences in changes developing in the two muscles during hypokinesia are attributable to some disturbances, thus far unidentified, of metabolic processes in the peripheral or central elements of the innervation system of these muscles.

## BIBLIOGRAPHY


1. Portugalov, V. V., and Ilyina-Kakueva, E. I. AEROSPACE MEDICINE, Vol 44, 1973, pp 764-768.

2. Portugalov, V. V.; Il'ina-Kakuyeva, Ye. I.; Rokhlenko, K. D.; et al. in: "Tezisy 3-y Vsesoyuznoy konferentsii po aviatsionnoy i kosmicheskoy meditsine" [Summaries of Papers Delivered at the 3d All-Union Conference on Aviation and Space Medicine], Moscow, Vol 2, 1969, pp 163-167.

3. Starostin, V. I.; Portugalov, V. V.; and Il'ina-Kakuyeva, Ye. I. DOKL. AN SSSR [Reports of the USSR Academy of Sciences], Vol 190, No 5, 1970, pp 1215-1217.

4. Dubowitz, V. "Developing and Diseased Muscle. A Histochemical Study," London, 1968.

5. Engel, W. K. NATURE, Vol 191, 1961, pp 389-390.

6. Resnick, J. S., and Engel, W. K. in: "Exploratory Concepts in Muscular Dystrophy and Related Disorders" (A. T. Milhorat, editor), Amsterdam, 1967, pp 255-267.

7. Duchen, L. W., and Strich, S. J. J. PHYSIOL., Vol 189, 1967, pp 2-4.

8. Duchen, L.W.; Excell, B. J.; Patel, R.; et al. J. NEUROL. SCI., Vol 21, 1974, pp 391-417.

9. Hoffman, H. AUST. J. EXP. BIOL. MED. SCI., Vol 28, Pt 3, 1950, pp 383-397.

UDC: 612.766.2.015.32

# EFFECT OF PHYSICAL LOADS ON SOME PARAMETERS OF LIPID AND CARBOHYDRATE METABOLISM DURING HYPOKINESIA

Moscow KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA in Russian
No 6, 1977 pp 36-40

[Article by T. M. Lobova and A. V. Chernyy, submitted 17 Apr 75]

[Text] The decreased amount of physical labor performed by most of the inhabitants of economically developed countries alters reactivity of the organism, causes progression of many diseases and complicates their course [1-7].

Physical exercise is the most accessible of measures directed toward prevention and elimination of the adverse consequences of hypokinesia. However, there is information indicative of diminished physical fitness and impairment of oxygen balance following such loads against the background of hypodynamia [8-10]. It is not known how soon and to what extent an organism, which has adapted to a minimal physical load, can adapt to intensified muscular exercise.

When assessing the effectiveness of physical loads in the case of restricted motor activity, one cannot fail to take into consideration the metabolism of carbohydrates and fats, which are the main energy resources used for muscular contractions. In this work, we tried to study some aspects of carbohydrate and lipid metabolism in the presence of hypokinesia and with the use, against this background, of lengthy physical loads.

## Methods

Experiments were conducted on 140 white, male rats weighing 150-230 g with different amounts of motor activity. The first group of animals was on an unrestricted regimen, in ordinary vivarium cages, with 5 animals in each; the second group was on an unrestricted regiment with measured physical exercise (swimming at 30-32° temperature) according to the method recommended by V. M. Pinchuk et al. [11]. The third group of animals was kept in small cages that restricted mobility; in the fourth group, hypokinesia was combined with swimming, following a regimen analogous to the one of the second group. The animals were decapitated on the 15th, 30th,

60th and 90th experimental days. Cholesterol of serum or whole blood was assayed by the method of Mrskos and Tovarek as modified by Zurkowski [12], nonesterified fatty acids (NEFA) were assayed according to Duncombe [13], ketone bodies (acetone + acetoacetic acid) according to Natelson [14], with some modifications; we assayed cholesterol of skeletal muscles and the liver by the colorimetric method of S. D. Balakhovskiy and I. S. Balakhovskiy [14] and total lipids, according to Huergo; glycogen was isolated from tissues according to Good et al. [16] and it was assayed according to Kemp and Kitz [17]. Lipolytic activity of fatty (epididymal) tissue was evaluated by a modification of the Gordon and Cherkes [18] method, expressing it in microequivalents of NEFA per milliliter per gram tissue.

Results and Discussion

As compared to the control, there was a 92% drop in glycogen content of the liver on the 15th day of restricted muscular activity, 62% drop in skeletal muscles, 90.4 and 18% drops, respectively on the 30th day; 93.5 and 53.3% on the 60th, 88.2 and 33.2% drops on the 90th day (Figures 1 and 2). Lipolysis in fatty tissue increased by 41.1% on the 15th day and remained on a stable high level; it increased by 31.9% on the 30th day and 39.4% on the 90th. The reliable elevation of blood NEFA and ketone body levels (by 66.6 and 282.4%, respectively, on the 15th day; 47.6 and 105.9% on the 30th day; 80.9 and 323.5% on the 60th day; 128.6 and 470% on the 90th day)(Figure 3), decrease in total lipid content of muscles (by 24.9% on the 15th day, 25.9% on the 30th and 30.1% on the 60th), as well as the tendency toward a decline of level thereof or reliable decline in the liver on the 15th, 30th and 60th days, are indicative of the rather high efficiency of lipolytic processes, not only in fatty, but other tissues. On the 90th day, the lipid level of the liver rose by 29.7%, apparently due to intensification of synthesis of triglycerides from fatty acids transported by blood. Accumulation of lipids and depletion of liver glycogen is one of the adverse manifestations of prolonged hypokinesia.

As can be seen from the submitted data, with marked reduction of motor activity there is depletion of carbohydrate resources of tissues, intensified mobilization and breakdown of fats, and fatty acids become intensively involved in oxidative processes. When they are dissociated, a considerable amount of acetyl-coenzyme A is formed, and it is converted into ketone bodies against the background of diminished carbohydrate metabolism (and this is confirmed by our results); it can also be involved in cholesterol synthesis. The elevation of cholesterol level in blood, skeletal muscles and liver of experimental animals, which is observed at all stages (by 25.9, 22.9 and 11.8%, respectively on the 15th day; 25, 27.7 and 14.5% on the 30th day; 18.3, 40.8 and 11.6% on the 60th day; 32, 24.4 and 31.7% on the 90th day) is most likely attributable to increased synthesis thereof in tissues.

In our opinion, the progressive increase in blood NEFA (with a maximum on the 90th day), with relatively stable high lipolysis in fatty tissue, is indicative not only of intensified breakdown of fat, but slower synthesis

thereof in the reservoir. This assumption appears to be quite founded, since there have been several reports [19-20] of slower re-esterification of fatty acids in fatty tissue when blood glucose level dropped and, according to our findings in this study, there is a tendency toward hypoglycemia and a severe reduction of glycogen resources in the presence of hypokinesia.

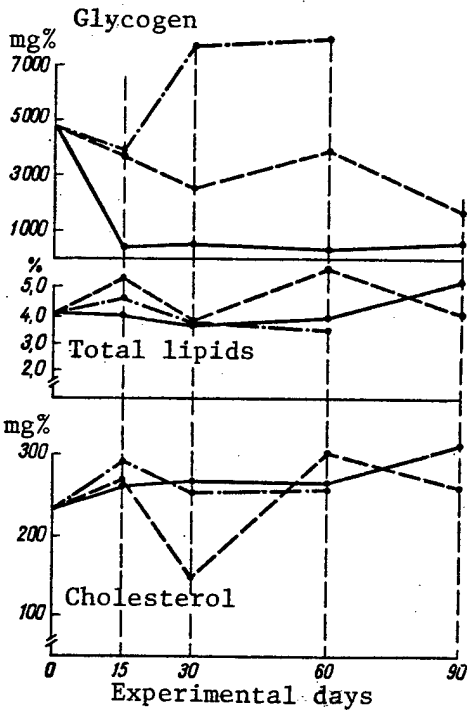



Figure 1.
Glycogen, total lipid and cholesterol levels in the rat liver as related to different regimens of motor activity.

Following applies to all figures: solid line, hypokinesia; dash line, hypokinesia alternating with exercise; dot and dash line, unrestricted regimen, alternating with exercise.

Here and in Figure 2: x-axis, duration of experiment; y-axis, glycogen (mg%), total lipid (%) and cholesterol (mg%) levels.

As can be seen in Figures 1-3, lipid metabolism underwent the most appreciable change at the early stages, namely the 15th day, in animals kept in a common cage, under the influence of swimming; and these changes were similar in several respects to those observed with hypokinesia. Thus, there was a significant elevation of NEFA (by 61.9%) and ketone body (205.9%) levels in rat blood, with increase in cholesterol content of the liver (by 24.1%) and skeletal muscles (17.2%), and decrease (21%) in glycogen content of tissues. Evidently, the same direction of changes in lipid and carbohydrate metabolism in animals kept on different regimens, with respect to muscular activity, reflects the common nonspecific reactions that occur in the course of adaptation to unusual situations, and it is related to increased activity of the hypophyseo-adrenal system.

On the 30th day, in the second group of animals, blood NEFA and ketone body levels, tissular cholesterol, total lipids of liver and glycogen of skeletal muscles were found to be on the same level as in the control, whereas liver glycogen level increased by 63%. This indicates that with gradually increasing loads the organism of healthy rats compensates relatively rapidly for the

increasing energy requirements by intensifying glycogen synthesis in the liver. On the 60th day, the level thereof in the liver is higher, as before (by 67%), than in the control, while in skeletal muscles it dropped reliably by 29%. This was associated with another increase in blood NEFA content by 45.2%) and an unreliable increase in ketone bodies (58.8%); total lipids of muscles were decreased, and there was a tendency toward decrease in level thereof in hepatic tissue (by 13.8%). Consequently, fats are utilized, along with carbohydrates, to maintain the energy balance in the presence of prolonged and increasing loads.

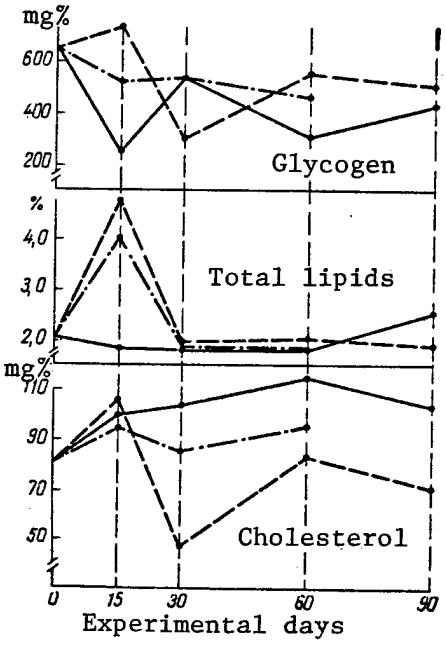



Figure 2.
Glycogen, total lipid and cholesterol content of rat skeletal muscles as related to different regimens of motor activity.

In rats whose mobility was limited (4th group), under the influence of swimming there was also an increase in lipolysis in fatty tissue; for example, there was a 40.2% increase on the 60th day, with the most marked elevation of NEFA level, as compared to other experimental groups: by 119% on the 15th day, 71.4% on the 30th and 95.2% on the 60th day. By the 90th day, the extent to which it exceeded the control, i.e., 42.9%, was relatively lower than in the third group of animals (128.6%). On the 30th and 60th days, there was a tendency toward decline of lipid levels, by 6.6 and 9.4%, respectively, in skeletal muscles; it became reliable on the 90th day (22.1%). There was no appreciable difference between lipid content of the liver on the 30th and 90th days, as compared to the control level, but on the 15th and 60th days it was 32.1 and 41.7% higher. Evidently, the intensified access of free fatty acids to the liver (a maximum level thereof in blood plasma was observed at the same times) is instrumental in accumulation of neutral fats in it and affects the level of total lipids, which is consistent with the data of N. N. Yakovlev [21], who observed deposition of fats in the liver with prolonged physical loads.

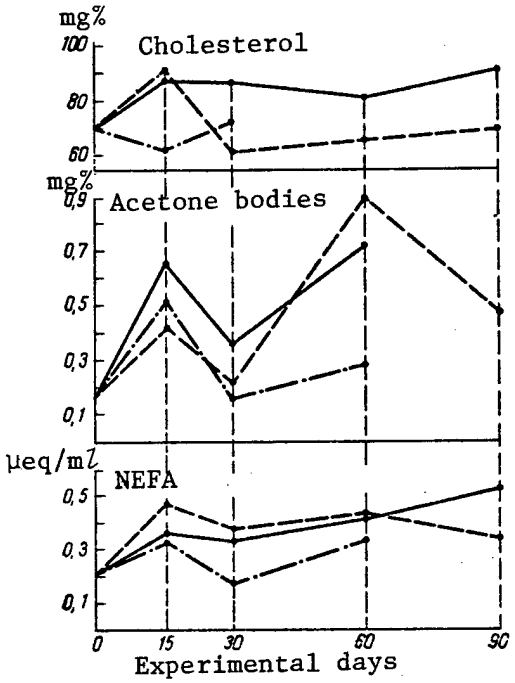



Figure 3.
Cholesterol, NEFA content of serum and acetone bodies (acetone + acetoacetic acid) in whole blood as related to various regimens of motor activity.

X-axis, duration of experiment; y-axis, cholesterol (mg%), acetone bodies (mg%) and NEFA (μeq/mℓ)

Unlike the rats with limited muscular activity, the animals kept under similar conditions but forced to swim presented a less significant elevation of blood ketone bodies (with the exception of the 60th day), namely, there was a 147% increase, as compared to the control, on the 15th day; 23.5% on the 30th and 170.6% on the 90th day. The glycogen content of tissues diminished to a lesser extent in the fourth group of rats: by 22% in the liver and the same as in the control on the 15th day; 46 and 55% decrease, respectively, on the 30th day; 17 and 15% on the 60th day; 66 and 21% on the 90th day. While it was possible to demonstrate a distinct elevation of blood and tissue cholesterol levels in the case of hypokinesia throughout the experimental period, against this background the cholesterol content of blood and skeletal muscles increased by 31.1 and 29.6%, respectively, only on the 15th day under the influence of physical exercise. Thereafter, its level in blood did not change appreciably, as compared to the control, while in skeletal muscles, on the contrary, it decreased by 42.1 and 15.1% on the 30th and 90th days, respectively. Cholesterol level of the liver rose by 13.3 and 26.3% on the 15th and 60th days, dropped by 36.9% on the 30th day and was the same as in the control on the 90th day.

Thus, in the presence of experimental hypokinesia and combination thereof with physical loads, the role played by fats increases with regard to maintaining the energy balance of the organism. The loads, which enhance mobilization and catabolism of lipids, aid in fuller utilization thereof and prevent accumulation of ketone bodies in blood, cholesterol in blood and

skeletal muscles, thereby retarding but not completely preventing, under our conditions, some of the adverse effects of hypokinesia. The volume and duration of such loads must be strictly regulated, since an excessive load could intensify triglyceride synthesis, as a result of depletion of liver glycogen and increased access of fatty acids, and this creates a hazard to normal liver function.

## BIBLIOGRAPHY


1. Korobkov, A. V. in: "Problemy sportivnoy meditsiny" [Problems of Athletic Medicine], Moscow, 1965, p 133.

2. Kryachko, I. A. Ibid, p 136.

3. Parin, V. V. VESTN. AMN SSSR [Vestnik of the USSR Academy of Medical Sciences], No 6, 1965, p 5.

4. Acheson, R. M. YALE J. BIOL. MED., Vol 35, 1962, p 143.

5. Malhotra, S. Z. BRIT. HEART J., Vol 29, 1967, p 895.

6. Tailor, H. J.; Henschel, A.; Erickson, L.; et al. FED PROC., 4, 1, 1945, p 71.

7. Tailor, H. J.; Henschel, A.; Brozek, J.; et al. J. APPL. PHYSIOL., Vol 25, 1949, p 223.

8. Kakurin, L. I. KOSMICHESKAYA BIOL. [Space Biology], No 2, 1968, p 59.

9. Katkovskiy, B. S.; Machinskiy, G. V.; Toman, P. S.; et al. Ibid, No 4, 1974, p 43.

10. Kovalenko, Ye. A.; Galushko, Yu. S.; Sherashev, S. G.; et al. Ibid, No 1, 1975, p 13.

11. Pinchuk, V. M.; Levina, L. I.; and Popov, V. N. BYULL. EKSPER. BIOL. [Bulletin of Experimental Biology], No 5, 1973, p 18.

12. Zurkowski, R. in: "Mikrometody v klinicheskoy laboratorii" [Micromethods in Clinical Laboratory Practice], edited by A. V. Karakashova and Ye. P. Vichev, Sofia, 1968, p 208.

13. Duncombe, W. G. CLIN. CHIM. ACTA, Vol 9, 1964, p 122.

14. Natelson, S. "Microtechniques of Clinical Chemistry," Springfield, 1957.

15. Balakhovskiy, S. D., and Balakhovskiy, I. S. in: "Metody khimicheskogo analiza krovi" [Methods of Chemical Analysis of Blood], Moscow, 1953, p 483.

16. Good, C. A.; Kramer, H.; and Somogyi, M. J. BIOL. CHEM., Vol 100, 1933, p 485.

17. Kemp, A., and Kitz, A. J. M. BIOCHEM. J., Vol 56, 1954, p 646.

18. Gordon, R. S., and Cherkes, A. PROC. SOC. EXP. BIOL. (New York), Vol 97, 1958, p 150.

19. Vasil'yeva, Ye. D. USPEKHI SOVR. BIOL. [Advances in Modern Biology], Vol 78, Vyp 1(4), 1974, p 139.

20. Davtyan, N. K. PROBL. ENDOKRINOL. [Problems of Endocrinology], No 6, 1963, p 33.

21. Yakovlev, N. N. FIZIOL. ZH. SSSR [Physiological Journal of the USSR], No 3, 1952, p 332.

UDC: 612.826.4+612.432+612.451]"52"

# CIRCADIAN CHANGES IN ACTIVITY OF THE HYPOTHALAMUS-HYPOPHYSIS-ADRENAL SYSTEM IN ANIMALS DIFFERING IN INDIVIDUAL RADIOSENSITIVITY

Moscow KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA in Russian
No 6, 1977 pp 40-45

[Article by Yu.P, Druzhinin, Ye. I. Zubkova-Mikhaylova and G. N. Podluzhnaya,
submitted 10 Nov 75]

[Text] The question of individual radiosensitivity is one of the most complex ones in radiobiology. However, by now several methods have been refined for predicting individual radiosensitivity, according to the initial condition of the organism and its reactivity [1, 2]. When work on this problem began, it was believed that the individual differences in radiosensitivity to acute radiation are rather stable in time, although most experimenters preferred to expose animals to radiation as soon as possible after examining them in order to demonstrate correlations. When the interval between examination of a specimen and subsequent irradiation thereof was increased, the prognostic value of the proposed methods diminished sharply. After demonstration and investigation of the existence of circadian fluctuations in radiosensitivity [3, 4], one would have thought that, in spite of the changing "absolute" individual radiosensitivity in time, the difference between specimens would remain constant. True, a scrutiny of data referable to biorhtyhmological studies [5, 6] revealed that the variability of most parameters changes in the course of a 24-hour period. One would also expect that individual radiosensitivity of different specimens also varies asynchronously in the course of a 24-hour period. Moreover, it was noted that the range of individual variations of radiosensitivity diminishes if exposure to ionizing radiation was extended. This was particularly manifested in the case of exposure of rats, that were kept on a strict regimen with respect to lighting, to radiation for 24 h [7]. The index of individual variations ($I = \frac{LD_{84/30}-LD_{16/30}}{LD_{50/30}}$) decreased by 3-6 times. The above facts warranted the assumption that the individual differences in radiosensitivity of different specimens are unstable as function of time, they can decrease or increase, depending on the time (of day) of examination [7]. In addition, it could have been expected that some specimens could be referred to either the radioresistant or radiosensitive category, depending on the time of day, in view of individual differences in both phase and amplitude of circadian

processes that determine radiosensitivity. This hypothesis is confirmed by a number of indirect data. At the same time, it is known that activity of the hypothalamus-hypophysis-adrenal (HHA) system is subject to daily fluctuations [8, 9]. And since the daily changes in radiosensitivity and state of systems of the organism that are critical for radiation are related to viariations in activity of the hypophyseoadrenal system [10], the task was undertaken of determining the activity of the HHA system at different times of day in specimens differing in sensitivity to acute radiation. We used one of the most effective prognostication methods, the epinephrine test, to select two groups of animals differing in radiosensitivity [7].

## Methods

These studies were conducted on male Wistar rats weighing 180-200 g, that were kept in groups of 5-6 animals in a cage with provision of 12 h of light and 12 h of darkness (the lights were turned on at 0400 hours). The animals were examined at 0900-1200 hours to evaluate individual radiosensitivity. For this purpose, we assayed peripheral blood leukocytes before and 1 h after intraperitoneal injection of epinephrine (0.25 mg/kg weight). Rats which demonstrated a decrease by more than 4000 leukocytes per $mm^3$ in the epinephrine test were classified as radioresistant; they were equivalent in radiosensitivity to animals in whom a high leukocyte reaction was observed in the epinephrine test, $\Delta L>5000$. Rats with changes in number of leukocytes not exceeding 1000 in either direction were classified as radiosensitive. Thus, of the 300 rats, we selected 40 radioresistant and 40 radiosensitive specimens. We did not then regroup the animals in the cages. We sacrificed 5 animals from each group every 3 h for 24 h in order to evaluate the daily changes in condition of the HHA system. We used Bouin fluid and 10% formalin as fixing agents; neurosecretion was demonstrated with fuchsin paraldehyde by the method of Gomori or Gab with additional hematoxylin staining according to Mayer. Visual estimation of secretion in the hypothalamus-hypophysis system was supplemented by measurement of the large diameter of neuronal nuclei. To determine the functional activity of the adrenal, we took into consideration the weight of the organ, area of cortex and different zones thereof, diameter of nuclei of glomerular cells, lipid and ketosteroid content of cortical zones. Lipids were demonstrated with Sudan III and Sudan IV, ketosteroids were demonstrated with Schiff reagent. We assayed 11-OCS [hydroxycorticosteroids] fluorimetrically by the method of I. Ya. Usvatova and Yu. A. Pankov [11].

## Results and Discussion

In the morning (0500-0800 hours), the condition of large-cell nuclei of the hypothalamus in rats of the radiosensitive group were varied. The level of secretion was low in the paraventricular nucleus, whereas activity of neurons of the supraoptical nucleus was on a higher level. The neurons of this nucleus were large and clear; the secretory granules in the processes were demonstrable in small quantity both in the region of the nucleus and along the course of the hypothalamo-hypophyseal tract. At 1100 hours, there was prevalence of cells containing secretory granules in the perinuclear

region of the paraventricular nucleus; excretion of secretions in the processes was demonstrable. In the supraoptical nucleus, clear neurons were retained; there were also small, dark (dehydrated) cells. The neurohypophysis had a low secretion content. Progressive intensification of synthesis of secretions in neurons of the paraventricular nucleus continued at 1400 hours; there was also intensification of the process of migration of secretions in the processes. Conversely, the secretion level dropped in the supraoptical nucleus. There was an increase in amount of secretory material in the median eminence and posterior lobe of the hypophysis.

At 1700 hours, there was a high level of neurosecretions in the hypothalamopituitary system. There was accumulation of neurons in the paraventricular nucleus; their bodies and processes contained a large amount of secretions. There was no appreciable change in conditions of neurons in the supraoptical nucleus. There was a large amount of secretions, in the form of fine granulation and neurosecretory dilatations varying in diameter, in the neurohypophysis. There was the impression that there was stasis of secretory material in the system due to diminished mobilization.

At 2000 hours, the functional activity of macrocellular nuclei increased to varying extents in this series of animals. In some rats, there was intensification of the process of migration of secretion along the processes; in others, many cells were at the accumulation stage (Figure 1a). In the neurohypophysis, the secretion is concentrated in the large Herring bodies and neurosecretory dilatations (Figure 1b and 1c). Thereafter (2300-0200 hours), the process of migration of secretions along the axons was the most marked in the hypothalamohypophyseal system, while synthesis thereof gradually diminished. This was indicated by the presence of empty or small dark neurons in the nuclei, particularly the paraventricular, and large amounts of secretory granules along the course of the hypothalamohypophyseal tract. At 0200 hours, neurosecretory synthesis was demonstrable only in the supraoptical nucleus.

In the radioresistant group of rats, the circadian activity of the hypothalamohypophyseal system was characterized by certain distinctions. At 0500 hours, there was moderate secretory synthesis in the bodies of the neurons, as well as migration thereof along the process in macrocellular nuclei. Small, dark cells were encountered in a negligible quantity. There was a low level of secretions in the neurohypophysis. At 0800 hours, most of the neurons of the paraventricular nucleus were in the resting phase, whereas the cells of the supraoptical nucleus remained active. There was no change in amount of secretions in the neurohypophysis. From 1100 to 2000 hours, there was a gradual increase in activity of secretory neurons. At the start of this period, there was prevalence of the process of secretion with signs of accumulation of secretions, which was the most marked at 1700 hours. At 2000 hours there was intensified migration of secretions along the processes, and this occurred both in the region of the nucleus and in the hypothalamohypophyseal tract (Figure 1d and 1e). The posterior lobe contained a moderate amount of secretory material, which could be the result of rather intensive migration of active elements into the blood stream (Figure 1f). At

2300 and 0200 hours, there was gradual decrease in synthesis, whereas migration of secretions to the endings was more intensive, and the bodies of neurons became free of them.

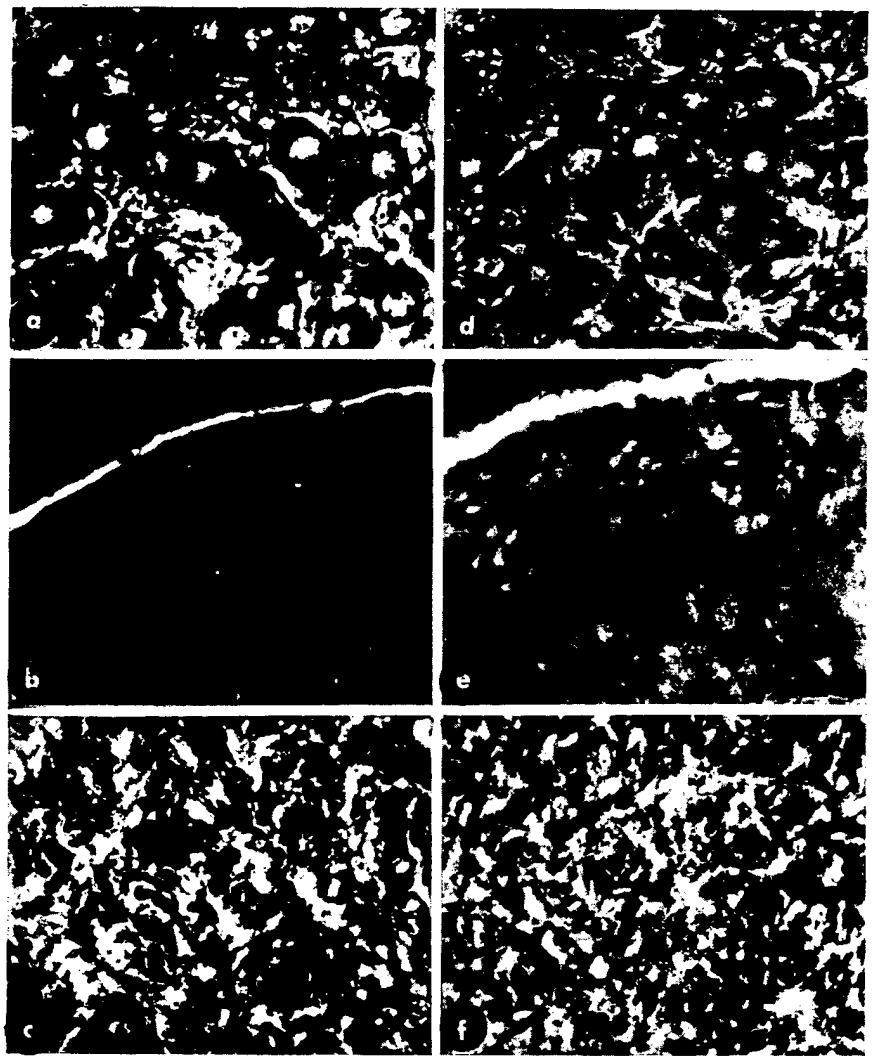



Figure 1. State of Wistar rat neurosecretory system at 2000 hours

a) paraventricular nucleus
b) median eminence
c) posterior pituitary lobe of radiosensitive rats
d) paraventricular nucleus
e) median eminence
f) posterior pituitary lobe of radioresistant specimens

Fuchsin paraldehyde stain according to Gomori

Magnification:
a, d) ocular 10×; objective 40×
b-c, e-f) ocular 10×; objective 20×

The differences in neurosecretory activity between radioresistant and radiosensitive rats were distinctly demonstrable when we measured the mean diameter of neuronal nuclei at each time that we sacrificed animals (Figure 2). In radiosensitive rats, the mean diameter of nuclei of supraoptical nucleus neurons changed insignificantly over a 24-h period and presented 2 peaks, at 1400 and 2000 hours. In radioresistant rats, the diameter of neuronal nuclei in the two macrocellular nuclei fluctuated synchronously and presented one reliable increase at 2000 hours. It is also interesting to mention that the absolute size of diameters of neuronal nuclei was smaller in this group of rats, whereas secretory activity was more intensive in neurons, there being no cells in the phase of profound rest (dark cells).

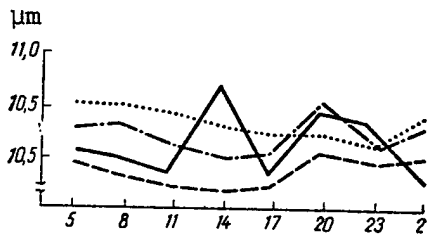



Figure 2.
Mean diameters of neuronal nuclei of macrocellular nuclei at different times of day in radiosensitive (solid line and dotted line) and radioresistant (dash-dot and dash lines) specimens. Solid and dotted lines--paraventricular nucleus; dash-dot and dash lines, supraoptical nucleus

Analysis of morphometric indices characterizing adrenocortical function leads to the conclusion that it presents dissimilar circadian fluctuations in radiosensitive and radioresistant rats. Thus, in radiosensitive animals, the weight of the adrenal increases twice, at 1400 hours (19 mg%) and 2300 hours (18 mg%), after its minimum level at 0800 hours (16 mg%). The area of the cortex also increased twice, at 1400 hours and to a lesser extent at 0200 hours, and the changes were referable mainly to the area of the fascicular and reticular zones; the range of fluctuation in area of these zones was 3.4 to 4.2 $cm^2$.

We divided the diameter of cells of the fascicular zone into three categories at 10-unit intervals, and expressed the number of nuclei in each category as percentages. In radiosensitive rats, the diameter of the nuclei increased at 0800 and 1100 hours; there was a high percentage of nuclei with small diameter between 1400 and 2000 hours, and some shift at 2300 hours in the direction of increase in nuclear diameter (Figure 3). There was a correlation between changes in diameter of fascicular zone cell nuclei and lipid and ketosteroid content. Accumulation of lipids and ketosteroids was observed in the periods when there was an increased percentage of nuclei with small diameter (1400 and 1700 hours); conversely, an increase in nuclear diameter was combined with a decreased amount of lipids and ketosteroids in the fascicular zone (1100 and 0200 hours). Figure 4 illustrates the 11-HCS content of peripheral blood, the level of which reached a maximum at 1400 hours in this group of animals, then gradually declined.

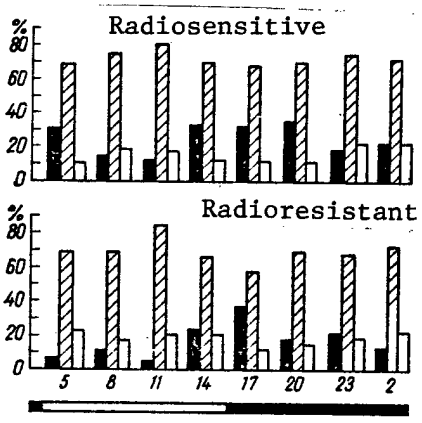



Figure 3.
Percentage of 3 categories of cell nucleus diameters in adrenal fascicular zone. Black columns, 1st category; striped, 2d; white, 3d.

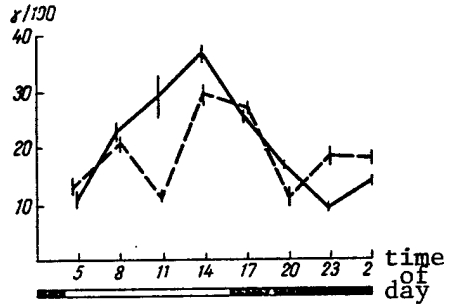



Figure 4.
Daily fluctuations in 11-HCS content of peripheral blood of radiosensitive (solid line) and radioresistant (dash line) rats

The changes were different in all of the above indices over a 24-h period in the radioresistant animals. The weight of the adrenals dropped to 15 mg% between 0500 and 0800 hours; it reached 17 mg% at 1400 hours staying on a plateau that lasted to 2300 hours. The area of cortical substance and of its different zones underwent negligible change over the 24-h period; the range of fluctuations of area of the fascicular and reticular zones, 3.2-3.7 $cm^2$, was unreliable. The diameter of cell nuclei of the fascicular zone increased at 1100 hours, decreased significantly at 1700 hours and again increased at 0200 and 0500 hours (see Figure 3). A high lipid and ketosteroid level was observed only at 0800 hours, it dropped significantly by 1100 hours and did not change appreciably thereafter. The 11-HCS level of peripheral blood rose twice in the course of a 24-h period (see Figure 4).

Evidently, all of the studied indices of functional activity of the adrenal cortex of radioresistant rats present a lower amplitude of circadian fluctuations than analogous indices of radiosensitive rats.

Thus, the facts we demonstrated confirm the hypothesis that animals differing in reaction to acute radiation are characterized by dissimilar course of periodic circadian processes. These differences in amplitude, position of acrophases and form of periodic process were noted both on the level of the system that is critical for radiation lesion, the hemopoietic system, and in activity of the main central clock, the HHA system.

It should be noted that radioresistant animals, with lower amplitude of changes in the HHA system, were characterized by considerably wider amplitude of fluctuations in effector elements. Thus, the amplitude of daily changes in leukocyte content of peripheral blood of radioresistant and radiosensitive rats constituted 12,220 and 7690/$mm^2$, respectively, and the amplitude of

fluctuations of eosinophils constituted 453 and 119 cells/mm$^2$ [7]. The higher sensitivity of effector systems of radioresistant specimens to mediators and hormones is indicative of greater refinement of their homeostatic mechanisms.

The dissimilar course of circadian processes in some animals is probably due to the hiearchic correlations in the group. In our case, there were 5-6 specimens per cage. At the same time, among the tested animals, 16-20% (or 1 our of 5-6 rats) were radioresistant specimens. One would think that expressly these rats are the leaders of the groups.

BIBLIOGRAPHY


1. Pospisil, M., and Ueno, J. FOLIA BIOL. (Prague), Vol 10, 1964, pp 232-238.

2. Karpfel, Z., and Soska, J. Ibid, Vol 5, 1959, p 440.

3. Kuznetsova, S. S. in: "Voprosy obshchey radiobiologii" [Problems of General Radiobiology], Moscow, 1971, pp 180-190.

4. Pizzarello, O. J.; Witeofski, R. Z.; and Lyons, E. A. SCIENCE, Vol 139, 1963, pp 349-352.

5. DeKursi, P. in: "Biologicheskiye chasy" [The Biological Clock], Moscow, 1964, pp 91-103.

6. Sollberger, A. "Studies of Temporal Variations in Biological Variates," Stockholm, 1960.

7. Druzhinin, Yu. P.; Grigor'yev, Yu. G.; Podluzhnaya, G. N.; et al. in: "Funktsiya organizma i faktory kosmicheskogo poleta" [Functions of the Organism and Space Flight Factors], Moscow, 1974, pp 125-131.

8. Zubkova-Mikhaylova, Ye. I.; Polyakov, N. D.; and Ponomarenko, L. M. in: "Materialy 1-y Vsesoyuznoy konferentsii po neyroendokrinologii" [Proceedings of First All-Union Conference on Neuroendocrinology], Leningrad, 1974, p 65.

9. Popovichenko, N. V. "The Role of the Hypothalamic Neurosecretory System in Adaptive Reactions of the Organism," Kiev, 1973.

10. Fochem, K.; Michalica, W.; and Picha, E. STRAHLENTHERAPIE, Vol 133, 1967, pp 256-261.

11. Usvatova, I. Ya., and Pankov, Yu. A. in: "Sovremennyye metody opredeleniya steroidnykh gormonov v biologicheskikh zhidkostyakh" [Modern Methods of Assaying Steroid Hormones in Biological Fluids], Moscow, 1968, pp 38-48.

UDC: 612.35.014.477-063

# FUNCTIONAL STATE OF THE RAT LIVER UNDER THE PRIMARY DELETERIOUS EFFECT OF IMPACT ACCELERATIONS

Moscow KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA in Russian
No 6, 1977 pp 45-50

[Article by Ye. Ye. Simonov and K. B. Ryklin, submitted 24 Mar 76]

[Text] As we know, the primary damaging effect of impact accelerations involved in landings are manifested primarily in the form of microtrauma to lung tissue [1-3]. However, there are some facts, and first of all the nature of changes observed in biochemical composition of blood [4, 5], that suggest the involvement, to some extent, of other internal organs as well, in particular the liver. In order to settle this question, we conducted a parallel study of histology and histochemistry of the liver of animals exposed the impact accelerations of landing that did and did not elicit primary damage in the lungs. Along with the histological and histochemical study of hepatic tissue of experimental animals, we also investigated the biochemical composition of blood.

## Methods

Experiments were conducted on 184 male albino rats weighing 150 to 300 g, 40 of which served as a biological control, while the rest were divided into two almost equal experimental groups and exposed to impact accelerations in the "back-chest" direction on a special stand [1, 5]. The landing rate constituted 3 m/s and accelerations constituted 410±50 units [U] in the first experimental group, 10 m/s and 465±50 U, respectively, in the second.

All of the tests (histological, histochemical and biochemical) were performed at the same times: 4, 24, 48, 72 and 120 h after exposure. The data obtained were compared to the findings of tests performed before the experiment, or to the data on the control group of animals.

We evaluated the effects of accelerations on the basis of presence of lesions to the viscera, as established at autopsy and subsequent microscopic examination of tissue specimens.

Pinpoint effusions of blood in lung tissue served as an indication of the primary damaging effects of accelerations on the organism.

The animals were decapitated. The blood was collected, while the serum obtained upon retraction of clots was used to assay enzyme activity: aspartic and alanine aminotransferases (GAST and GALT), aldolase (ALD), lactate dehydrogenase (LDH) and nonspecific cholinesterase (NCE). In addition, we conducted dynamic observation of 10 rats in each experimental group, assaying cholesterol, urea and creatinine levels in blood. Blood was taken after making a small cut in the tip of the tail.

Enzyme activity was determined using the methods indicated in [5]; cholesterol, urea and creatinine were assayed by means of complex microchemical analysis [6].

For the histological and histochemical studies, we used the combined block technique, as well as hematoxylin-eosin stain according to Nissl, Shabadash reaction for glycogen, the method of Nachlas and Seligman for demonstration of succinate dehydrogenase (SDH), the Gomori method for α-glycerophosphate dehydrogenase (GPDH), NAD diaphorase and alkaline phosphatase (AP), the Glinner method for monamine oxidase (MAO) and Brachet method for RNA. Lipid content was determined by means of staining with scarlet red.

Results and Discussion

Macroscopic and microscopic examination of tissues of experimental animals revealed pathomorphological changes in the viscera only in the second group of rats. In this case, the lesions were localized primarily in the lungs which is typical, according to the literature [1, 2, 7], for the primary deleterious effects of accelerations involved in landing. As a rule, these were circumscribed effusions (1 to 3-8 mm in diameter) at the sites of which pneumonia developed in some cases. Effusions of blood were demonstrated in the liver of only two rats.

Histochemical examination of the liver revealed that the first group of animals developed negligible and very transient changes: minor drop of glycogen level in hepatic cells, some increase in AP activity, appearance of fine drops of lipid in some groups of cellular elements. There was no increase in concentration of RNA. There was a negligible increase in SDH activity only on the 3d-5th day after exposure. There was no change at all in activity of GPDH and MAO. In the second group of rats, the histochemical changes in the liver were more marked and consisted of appreciable and rapid (within 4-24 h) increase in activity of AP, SDH, MAO, RNA, decrease in GPDH activity, marked decrease in glycogen content, particularly in the cells of central segments of the lobes, and development of fatty dystrophy at the end of the 3d day. There was an appreciable decrease in NAD diaphorase activity in both experimental groups.

Blood tests (serum) established (see Table) that there was no change in most of the indices determined in the first group of animals, not counting the brief and minor (15-30%, as compared to the control) elevation of activity of GAST, GALT, ALD and LDH, observed primarily after 24 h, rather than immediately after exposure. The findings were different in the second group of animals. In this case, cholesterol and urea levels dropped appreciably (on some days, by 20-55%). There was a more appreciable increase in activity of GAST, GALT, ALD and LDH. The increment of activity constituted 20-40%. However, the most significant factor was the rapid appearance (within 4 h) and longer duration (up to 72 h), rather than quantitative aspect of the increase in activity. Moreover, the animals of the second group also presented some increase in NCE activity.

Analysis of the results of histochemical and histological studies of the first group of animals warrants the conclusion that the changes in some histochemical indices, unrelated to impairment of hepatic tissue structure, could be attributed to the functional category and reflect a general defense reaction in response to stimulation, which occurred in the course of preparation and performance of experiments. Evidently this is also indicated by the results of biochemical blood tests, according to which no changes were demonstrated in cholesterol, urea, creatinine and NCE synthesized in hepatic tissue of animals in the first group [9-11]. The lack of changes in these indices indicates that, in this instance, there were no disturbances in the relevant specialized functions of the liver. The brief and minor increase in activity of GAST, GALT, ALD and LDH, recorded only 1 day after impact accelerations, rather than immediately, was probably due to a change in hormonal status, in particular, intensified production of corticosteroids and other hormones capable of stimulating the activity of certain enzymatic systems [10-13].

Albino rats are notable for high excitability, and they react readily to any stimuli. Their rapid development of a tension reaction was demonstrated in a technically exquisite study [14]. This author demonstrated an appreciable elevation of corticosterone level in blood of rats, even if they were simply picked up. The same reaction was observed under the influence of other stimuli, for example, sonic ones.

As for the histochemical changes in the liver of rats in the second group, they could apparently reflect some sort of discrete, possibly submicroscopic changes in structure of hepatocytes, as well as metabolic disturbances in liver cells, which is consistent with the results of biochemical blood tests.

If we consider these data from the standpoint of current conceptions of biochemical manifestations of mechanical trauma [15, 16], we cannot fail to arrive at the conclusion that the above-mentioned changes in biochemistry of blood in the second group of animals are the result of metabolic disturbances in the parenchyma of the liver.

Biochemical changes in rat blood after exposure to impact accelerations (M±m)

| Index | Experimental conditions | | Mean and range of fluctuation of initial state or control | Mean levels at different intervals (hours) after exposure (% of initial levels) | | | | |
|---|---|---|---|---|---|---|---|---|
| | descent rate, m/s | accel. M±m, units | | 4 | 24 | 48 | 72 | 120 |
| Cholesterol, mg% | 3 | 410±50 | 112 (10) 105—112 | 96 | 96 | 98 | 100 | 100 |
| | 10 | 465±50 | 96 (10) 88—105 | 100 | 94 | 90 | 80 | 87 |
| Urea, mg% | 3 | 410±50 | 21 (10) 12—32 | 104 | 95 | 98 | 104 | 100 |
| | 10 | 465±50 | 24 (10) 18—26 | 93 | 78 | 85 | 91 | 106 |
| Creatinine, mg% | 3 | 410±50 | 1,63 (10) 1,2—2,2 | 102 | 103 | 100 | 97 | 102 |
| | 10 | 465±50 | 1,45 (10) 1,0—2,2 | 130 | 130 | 115 | 143 | 100 |
| GAST | 3 | 410±50 | 215±7 (40) | 107 (6) | 116 (10) | — | 95 | — |
| | 10 | 465±50 | 215±7 (40) | 120 (8) | 135 (16) | — | 107 (12) | — (12) |
| GALT | 3 | 410±50 | 100±4 (40) | 90 (6) | 115 (10) | — | 75 (6) | |
| | 10 | 465±50 | 100±4 (40) | 100 (8) | 130 (16) | — | 130 (12) | |
| ALD | 3 | 410±50 | 630±30 (40) | 100 (6) | 133 (10) | — | 127 (6) | |
| | 10 | 465±50 | 630±30 (40) | 127 (8) | 127 (16) | — | 110 (12) | |
| LDH | 3 | 410±50 | 2900±300 (40) | 66 (6) | 124 (10) | — | 96 (6) | |
| | 10 | 465±50 | 2900±300 (40) | 66 (8) | 140 (16) | — | 96 (12) | |
| NCE | 3 | 410±50 | 0,56±0,01 (30) | 95 (6) | 104 (10) | — | 102 (6) | |
| | 10 | 465±50 | 0,56±0,01 (30) | 93 (8) | 110 (16) | — | 130 (12) | |

Note: In the control, enzyme activity was expressed in the following units: GAST, GALT and LDH, in μg pyruvic acid per mℓ serum; ALD, in units of optical density × 100 × serum dilution; NCE, in pH units. The number of animals is given in parentheses.

According to data submitted in several works [15, 16], in the case of trauma, particularly when it leads to significant impairment of tissular integrity (closed fractures with displacement of fragments, compound fractures of bones of the lower extremities, extensive crushing of muscles, etc.), there is elevation of total nitrogen level in blood, with quantitative prevalence of urea nitrogen, which is related to local (in the traumatized area) and generalized breakdown of proteins. This is the so-called catabolic phase of systemic metabolic reaction of the organism to trauma, which is indicative of good adaptation to the deleterious factor. At the same time, instances have been encountered when the victim's blood presented a low urea content and high levels of other intermediate and end products of nitrogen metabolism, which is evaluated as an indication of the patient's poor condition. Such states are also characterized by disturbances referable to deamination, synthesis of urea and plasma proteins, hypocholesteroemia, impaired carbohydrate metabolism, ketonemia, etc., which is indicative of impairment of various specialized functions of the liver. It is important to mention that impairment of partial liver functions is not infrequently demonstrated in the presence of cerebrocranial trauma, which is indicative of the secondary nature of functional disorders [17-19].

If we compare this information to the results of dynamic assays of blood cholesterol, urea and creatinine in the second group of animals, we shall become readily convinced of the similarity thereof. This circumstance warrants the belief that, in this instance, the changes in cholesterol and urea levels are indeed a reflection of disturbances referable to the corresponding partial functions of the liver: biosynthesis of cholesterol or its esters and biosynthesis of urea. The elevation of creatinine level should apparently be interpreted as an indication of an intensive catabolic phase of the generalized metabolic reaction to trauma, characterized by prevalence of processes of protein breakdown over synthesis; in turn, the increase in enzyme activity in blood, which reflects the protein biosynthesizing function of the liver [8, 15], as well as the condition of cell membranes [23, 24], are an indication of disturbances of protein metabolism, on the one hand, and an indication of extent of increase in membrane permeability and, consequently, extent of "drain" of enzymes from cells, on the other. Since all of the enzymes we assayed are synthesized and present in significant quantities in the liver, the rapid increment in activity thereof in blood serum is primarily indicative of lesion to expressly this organ.

Thus, the results of this investigation warrant the conclusion that not only lung tissue, but parenchyma of the liver are affected by impact accelerations of landing, which induce a "primary deleterious effect" in the form of microtrauma (focal effusions of blood). However, unlike injuries localized in lung tissue, those involving the parenchyma of the liver cannot be visually identified, since they are limited primarily to changes in metabolic processes. The distinctive changes in biochemistry of blood are an indication of these disturbances; as we have seen, they are indicative of distortion or depression of some specialized functions of the liver: biosynthesis of cholesterol, biosynthesis of urea, etc. Hence, investigation of these functions is a promising methodological procedure for evaluation of the

intensity of an effect, mainly for in vivo differentiation between the injurious and noninjurious effects of impact accelerations occurring with landings.

## BIBLIOGRAPHY


1. Mirolyubov, G. P. BIOFIZIKA [Biophysics], No 2, 1966, pp 368-371.
2. Lalli, G., and Paolucci, G. RIV. MED. AERONAUT., Vol 29, 1966, pp 412-426.
3. Rushmer, R. F.; Green, E. L.; and Kingsley, H. D. J. AVIATION MED., Vol 17, 1946, pp 511-525.
4. Simonov, Ye. Ye. KOSMICHESKAYA BIOL. [Space Biology], No 6, 1975, pp 8-14.
5. Simonov, Ye. Ye., and Korzhen'yants, V. A. Ibid, No 4, 1968, pp 38-41.
6. Balakhovskiy, I. S., and Natochin, Yu. V. "Metabolism Under the Extreme Conditions of Space Flights and Simulation Thereof," Moscow, 1973.
7. Pearl, W.; Balazs, T.; and Buyske, D. LIFE SCI., Vol 5, 1966, pp 67-74.
8. "Clinical Enzymology," Warsaw, 1966.
9. Todorov, Y. "Clinical Laboratory Tests in Pediatrics," Sofia, 1961.
10. Aliyevskaya, L. L. VOPR. MED. KHIMII [Problems of Medical Chemistry], No 8, 1965, pp 57-63.
11. Razina, L. G. Ibid, No 2, 1960, pp 136-145.
12. Yudayev, N. A. Ibid, No 6, pp 559-573.
13. Segal, H. L., and Kim, J. S. J. CELL COMP. PHYSIOL., Vol 66, 1965, pp 11-12.
14. Dlusskaya, I. G. FIZIOL. ZH. SSSR [Physiological Journal of the USSR], No 3, 1966, pp 286-290.
15. Merezhinskiy, M. F., and Cherkasova, L. S. "Fundamentals of Clinical Biochemistry," Moscow, 1965.
16. Cherkasova, L. S. "Biochemistry of Trauma," Minsk, 1957.
17. Boyeva, Ye. M. "Essays on Pathophysiology of Acute Closed Brain Trauma," Moscow, 1968.

18. Burgman, G. P.; Biryukova, L. F.; and Kuklina, A. S. VOPR. NEYROKHIR. [Problems of Neurosurgery], No 6, 1966, pp 18-22.

19. Leont'yeva, A. A., and Kalinina, G. V. in: "Biokhimicheskiye izmeneniya v organizme pri travme" [Biochemical Changes in the Organism Associated With Trauma], Moscow, 1959, pp 84-93.

20. Podil'chak, M. D. "Clinical Enzymology," Kiev, 1967.

21. Shastin, R. N. "Problems of Enzymology," Moscow, 1964.

UDC: 615.2/.3.036.8.07:616.859.1-092.9.001.57

# MODEL OF MOTION SICKNESS IN DOGS USED TO EVALUATE EFFICACY OF PHARMACOLOGICAL AGENTS

Moscow KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA in Russian No 6, 1977 pp 50-53]

[Article by L. A. Radkevich, submitted 13 Mar 75]

[Text] The control of motion sickness has become one of the serious tasks for modern medicine in view of development of aviation and space research. In particular, many drugs have been tested for this purpose. It has been established that a number of cholinolytics and some antihistamines are effective for certain forms of motion sickness [1]. However, the diversity of manifestations of vestibular disorders and individual sensitivity to pharmacological agents do not enable researchers to consider the problem of treatment of this symptom complex to be solved.

The lack of clearcut and accessible screening methods makes it difficult to search for effective pharmacological agents. There are limitations to drug screening directly on humans and this is not economical. Moreover, it is difficult to take into consideration individual tolerance of vestibular stimuli under various conditions and characteristics thereof.

Preliminary drug screening under laboratory conditions, on animals, is expedient. However, we know that such animals as guinea pigs, rats, rabbits, as well as monkeys, are not particularly affected by motion sickness, or else the symptoms thereof are diminished. Experiments on cats are feasible only with the use of anesthesia, and this affects autonomic reactions. The literature concerning dogs is contradictory, due to the use of various forms of stimuli of vestibular receptors (vertical rocking, swinging on a four-pole swing, etc.). In the opinion of many authors, the main difficulty lies in selection of animals sensitive to rocking [2-6]. Thus, in the case of linear accelerations (vertical displacement) only 10-15% of the dogs are found to be susceptible to motion sickness.

In spite of some reservations, we believe that this animal species is promising for use to investigate motion sickness. In the first place, the symptom complex is vividly manifested in the form of progressive vegetative disturbances, to the extent of vomiting, in dogs with high susceptibility to motion sickness. In the second place, we can work with them

without using anesthesia. In the third place, the side-effects of various pharmacological agents (in particular, cytostatics) are similar in dogs and man [7]. The authors report that dogs are more sensitive to the side-effects of drugs than monkeys.

In view of the foregoing, we undertook the task of creating conditions for inducing motion sickness in dogs, under which the occurring accelerations would affect as many as possible of the receptor elements of the vestibular system, thus inducing maximum accumulation of stimuli.

Methods

We conducted our studies on intact mongrel dogs of both sexes weighing up to 10 kg. In refining the mode of motion, we created the possibility of generation of Coriolis accelerations throughout the exposure period (they are the most effective labyrinthine stimulus).

The device used consisted of a cabin corresponding to the size of the animal, with transparent walls. The cabin was suspended vertically on spring-loaded cables. The way the cabin was secured allowed for generation of concurrent vertical and rotational movements at an attenuating amplitude. The mean frequency of vertical swinging constituted 1/s at an amplitude of 70 to 100 cm. Concurrent rotation of the cabin about its axis was effected at a mean rate of 1.7 rev/s. The angular rate of rotation varied from 0 to 2 rev/s over a 5-min period.

The animal was not immobilized in the cabin and assumed a comfortable position, usually lying on its abdomen or side. We evaluated the dog's condition visually.

Control swining did not last over 60 min. In the experiment, we selected dogs that presented distinct signs of vestibular disorders or vomiting during the control swinging.

We tested several agents on the selected group of animals susceptible to motion sickness: marezine, an antihistamine (50 mg/kg); tigan, bromotigan (100 mg/kg), as well as metachlorpromide (1 mg/kg), which are antiemetics; diphenidol (2 mg/kg), a cholinolytic, and a few other products. The agents were injected intramuscularly in saline, 1 h before the control swinging. There was a 2-3-day interval between trials of various products on the same animal. If a positive effect of the drug was observed, the animal was submitted to control rocking before testing another agent. Another agent was tested only when there was no after-effect from a previously tested one.

We did not use products of the scopolamine type, since the data of some authors [2, 3] indicate that no effect was demonstrated with administration thereof when dogs were rocked for up to 45 min.

Results and Discussion

The method we used increased markedly the percentage of animals susceptible to motion sickness. Virtually all of the dogs submitted to control motion were sensitive to some extent or other. A high susceptibility to motion sickness was demonstrated in 70% of the tested animals, according to autonomic manifestations. A small number of animals was rejected when screened to appraise pharmacological agents. However, as can be seen in the Table, this group of "resistant" animals also presented vegetative reactions that were not frequent or vividly marked.

Vegetative reactions of 2 groups of animals during 60-min motion

| Duration of rocking, min | First group (susceptible to motion sickness) | Second group (resistant to motion sickness) |
|---|---|---|
| 1 | Orienting reaction, whining, faster respiration, mild salivation | No visible reactions, the animal is calm |
| 2 | Profuse salivation; the animal is depressed, licks itself constantly, yawns, rapid, superficial respiration | The animal is calm |
| 5 | Profuse salivation, intensified peristalsis | The animal is calm, occasionally licks itself |
| 10 | Profuse salivation, defecation, urination; the animal lies on the floor of the cabin and does not react when called by name | Occasionally licks itself, yawns |
| 15 | Profuse foaming at the mouth, nasal secretions; the dog yawns, moans and is depressed | Licks itself; no salivation |
| 25 | Frequent vomiting. Rocking stopped | No marked vegetative reactions |
| 30 | | Negligible salivation; the dog licks itself |
| 40 | | The animal is in satisfactory condition, responds when called, no increase in salivation |
| 50 | | Mild salivation; the dog licks itself and yawns |
| 60 | | Mild salivation, no vomiting; the animal's condition is satisfactory. Rocking stopped |

The characteristics of vegetative reactions developing within 60 min in two groups of dogs, a) sensitive to rocking and b) resistant to vestibular stimuli, are listed in the Table.

We see, from this table, that distinct vegetative reactions, such as profuse salivation, yawning, defecation, urination, faster respiration and, finally, vomiting were observed in sensitive animals, from the very first minutes of motion. It should be noted that, in some dogs, the vegetative disturbances appeared within a short period of time, 10-15 min from the start of motion. The resistant dogs presented late reactions that were considerably milder.

Trials of the above-mentioned drugs in accordance with the method of screening them that we developed revealed the following: Diphenidol had a beneficial effect on 80% of the tested animals, and one administration (2 mg/kg) had an effect that lasted for 3 days. Marezine (effective in 32% of the animals) and metachlorpromide (25%) were less effective. Tigan, as well as bromotigan, were ineffective.

These studies warrant the conclusion that products in the cholinolytic group and, to a lesser extent, antihistamines have protective properties with regard to canine motion sickness. It is remarkable, as indicated above, that a similar effect was obtained in studies on humans with the use of these groups of products. This confirms the fact that dogs are quite suitable biological objects for preliminary screening of pharmacological agents that prevent motion sickness.

The method we developed for exposing animals to motion can be recommended for use in research and appraisal of pharmacological agents that prevent development of motion sickness.

## BIBLIOGRAPHY


1. Lukomskaya, N. Ya., and Nikol'skaya, M. I. "The Search for Drugs Against Motion Sickness," Leningrad, 1971.

2. Noble, R. L. CANAD. J. RES. SECT. E, Vol 23, 1945, pp 226-234.

3. Idem, Ibid, Vol 26, 1948, pp 283-294.

4. Chinn, H. J., and Plotnikoff, N. P. J. APPL. PHYSIOL., Vol 5, 1953, pp 392-394.

5. Milch, L. J.; Frankl, H. D.; and Renzi, A. A. Ibid, Vol 14, 1959, pp 245-246.

6. Cook, L., and Toner, J. J. J. PHARMACOL. EXP. THER., Vol 110, 1954, pp 12-17.

7. Schein, P. S.; Ruth, D.; et al. CLIN. PHARMACOL. THER., Vol 11, 1970, pp 3-40.

UDC: 615.9:66.062.786

# TOXICOLOGY OF 1-4-DIOXANE

Moscow KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA in Russian
No 6, 1977 pp 53-57

[Article by Z. I. Pilipyuk, G. M. Gorban', G. I. Solomin and A. I. Gorshunova, submitted 20 May 75]

[Text] 1-4-Dioxane is used widely in industry, particularly as a solvent in the production of polymer materials.

Our objective here was to investigate the toxicity of this agent in the case of intake by inhalation and resorption, as well as to determine its irritant effect on mucous membranes and the skin.

## Methods

This study was conducted on white rats, white mice and rabbits.

Experimental animals were exposed to dioxane fumes once, for 4 or 2 h. We tested its topical and irritant effects on rabbits. We applied the agent in liquid form, 0.5 mℓ, uniformly over the shaved surface of the back (4×5 cm) once or twice (within 10 days), or else one drop was applied to the conjunctival sac of the eye. Experiments dealing with penetration of dioxane through the intact skin into the organism were conducted on mice, testing it once or repeatedly (10 times) using the conventional techniques.

We tested the rate of absorption of dioxane in blood on rabbits. We applied 5 mℓ of this agent on the shaved surface of the animals, covered the area with an airtight cover, then took blood (5-10 mg) from the auricular vein 5, 10, 30, 60, 120 and 180 min later for quantitative analysis on a G-C-1c chromatograph with flame-ionization detector. We tested the effects of prolonged inhalation of dioxane fumes (on 160 male white rats) in 800-liter chambers for 90 successive days. The air in the chambers was exchanged 4 times per hour; air temperature was in the range of 20-23°C and relative humidity, 60 to 70%. The experimental animals were divided into four groups. The rats were exposed to dioxane in concentrations of 20.5±1.0 mg/m$^3$ (1st group), 4.6±0.3 mg/m$^3$ (2d group) and 0.54±0.05 mg/m$^3$ (3d group). The 4th group served as a control. We evaluated the effect of dioxane on liver

function according to the change in activity of aspartate aminotransferase (AST) and alanine aminotransferase (ALT) in blood serum [1], and the Quick-Pytel' test as modified by I. G. Stepanova [2]. Kidney function was studied on the basis of diuresis, total protein of urine [3], urine chlorides (method of Votochek) and phenol red load [4].

In addition we studied the dynamics of body weight and nervous system function by the chronaximetric method.

Results and Discussion

As a result of acute tests, we established the following parameters of toxicity of dioxane: $LC_{50}$ = 46 (42.2÷50.1) mg/l, $LC_{84}$ = 52 mg/l, $LC_{16}$ = 40 mg/l with 4-h inhalation by white rats; $LC_{50}$ = 65 (61.3÷68.2) mg/l, $LC_{84}$ = 69.5 mg/l and $LC_{16}$ = 61 mg/l with 2-h inhalation by white mice.

The studies revealed that dioxane does not elicit changes in the rabbit skin. Single and repeated application of 25 mg liquid to the mucous membranes of the eye revealed its irritant effects: restlessness of experimental animals, closure of eyelids, profuse lacrimation, as well as development of conjunctivitis with significant edema on the following data. These signs were observed for 3-4 days.

Single and repeated (10 times) 4-h application of the agent revealed that dioxane has resorptive action. In the case of repeated applications of dioxane, the animals died on the 1st-8th days. The clinical signs were characterized by discoordination of movements, impaired respiratory function and depressed condition. Animals that survived failed to differ from controls, with regard to behavior, on the 2d day after the experiment.

As can be seen in Table 1, dioxane was demonstrated in blood within 5 min. This indicates that it is capable of penetrating rather quickly through the intact skin. It was also noted that the concentration of the agent in blood dropped by the 3d h after application to the skin.

Table 1. Dioxane content of blood, μg/ml

| Rabbit No | Time after application of dioxane, min | | | | | |
|---|---|---|---|---|---|---|
| | 5 | 10 | 30 | 60 | 120 | 180 |
| 1 | 0,2 | 0,9 | 0,9 | 0,9 | 0,5 | 0,2 |
| 2 | — | — | 1,0 | 1,0 | 1,0 | 0,5 |
| 3 | — | — | 1,6 | 1,5 | 0,4 | 0,4 |

Prolonged exposure to dioxane failed to elicit any deviations in general condition of animals and their behavior, as compared to the control. Body weight of experimental rats dropped by only 10-20% 45 days after the start of exposure, but then began to rise gradually and did not differ from levels in the control group by the time exposure was stopped.

Table 2. Effects of dioxane on rat liver and kidney function

| Parameters studied | Group | Dioxane concentration, mg/m$^3$ | Exposure days | | | | Recovery period |
|---|---|---|---|---|---|---|---|
| | | | 20 | 40 | 60 | 90 | |
| Diuresis, mℓ/day | 1 | 20,5 | 4,1±1,07 | 5,5±1,06* | 11,1±0,94 | 9,2±1,23 | 7,5±1,38 |
| | 2 | 4,6 | 5,5±1,29 | 5,1±0,96* | 10,6±0,75 | 9,4±0,45 | — |
| | 3 | 0,54 | 9,3±1,05 | 11,1±0,75 | 11,7±0,79 | 10,4±0,38 | — |
| | 4 | — | 7,1±1,34 | 10,6±0,66 | 9,7±1,15 | 9,2±1,05 | 11,7±0,66 |
| Protein, % | 1 | 20,5 | 0,47±0,08* | 0,33±0,06 | 0,97±0,17* | 1,90±0,08* | 0,66±0,15 |
| | 2 | 4,6 | 0,18±0,05 | 0,45±0,15 | 0,79±0,12* | 0,49±0,07 | — |
| | 3 | 0,54 | 0,10±0,08* | 0,24±0,05 | 0,70±0,04* | 0,40±0,02 | — |
| | 4 | — | 0,22±0,04 | 0,36±0,17 | 0,33±0,07 | 0,51±0,10 | 0,47±0,07 |
| Chlorides, mg/mℓ | 1 | 20,5 | 1,07±0,13* | 0,61±0,99* | 0,78±0,08 | 2,30±0,24* | 0,85±0,05 |
| | 2 | 4,6 | 1,22±0,11* | 0,96±0,10 | 1,08±0,17 | 0,78±0,05 | — |
| | 3 | 0,54 | 0,95±0,11* | 0,68±0,09* | 1,01±0,12 | 0,77±0,10 | — |
| | 4 | — | 1,78±0,21 | 0,90±0,09 | 0,85±0,02 | 1,53±0,17 | 0,85±0,08 |
| AST, U/mℓ | 1 | 20,5 | 28,3±1,38 | 28,1±1,38 | 32,4±1,40* | 35,9±1,83* | — |
| | 2 | 4,6 | 28,5±0,69 | 28,3±0,61 | 19,1±2,22* | 34,2±2,41* | — |
| | 3 | 0,54 | 28,4±1,82 | 27,8±2,22 | 26,0±2,84 | 24,6±3,84 | — |
| | 4 | — | 28,5±0,79 | 28,3±0,61 | 28,3±0,98 | 28,1±2,5 | — |
| ALT, U/mℓ | 1 | 20,5 | 12,6±1,83 | 12,3±0,97 | 9,6±0,61 | 30,7±0,55* | — |
| | 2 | 4,6 | 12,8±1,83 | 13,2±1,66 | 11,8±2,26 | 15,0±2,92* | — |
| | 3 | 0,54 | 12,1±1,42 | 11,0±1,21 | 23,3±2,28* | 22,5±1,0* | — |
| | 4 | — | 12,4±1,0 | 11,8±0,77 | 11,1±1,01 | 14,1±1,42 | — |

* $P<0,05$.

Table 2 lists data on the effects of dioxane on liver and kidney function. One of the sensitive indicators of liver function is transaminase activity. These enzymes are synthesized in the liver and participate in glycogen synthesis from proteins by means of transamination. Increased activity thereof in blood is indicative of impaired function of this organ. A statistically reliable increase in AST activity was demonstrated in the 1st and 2d groups of animals on the 60th and 90th experimental days ($P<0.05$). In this same period, the 3d group of animals presented elevation of ALT activity ($P<0.001$), and these changes were significant ($P<0.001$) in the 1st group of animals, on the 90th day. Some increase in transaminase activity under the influence of lower concentrations of dioxane (0.5 $mg/m^3$) at the early stages may be related to increased passage of enzymes from injured liver cells into blood, or increased permeability of histohematic barriers in the case of inflammation of the liver [5, 6].

In addition, the first and second groups of animals were found to be less tolerant to hexenal. The anesthetic effect doubled in duration, as compared to control animals, which is indicative of diminished detoxification function of the liver.

Studies of renal function revealed a change in 24-h diuresis, protein and chloride levels in urine. We failed to demonstrate changes in elimination of dye. As can be seen in Table 2, there was a reliable decrease in urine output in the 1st and 2d groups of rats, on the 40th day ($P<0.01$ and $P<0.001$).

On the 60th day, protein content increased ($P<0.01$, $P<0.02$ and $P<0.001$) in all experimental groups. At the end of the exposure period, these changes were more marked in the 1st group ($P<0.001$), which may be indicative of impaired renal filtration. Urine chloride content dropped on the 20th day in the 1st, 2d and 3d groups of animals ($P<0.02$, $P<0.01$ and $P<0.05$), and on the 40th day in the 1st and 3d groups ($P<0.05$ and $P<0.05$); a reliable increase ($P<0.02$) was observed only in the 1st group toward the end of the exposure period. Recovery occurred 10 days after exposure.

According to the data in the literature [4], the above changes in renal function are related to diminished rate of glomerular filtration, more intensive reabsorption of chlorides (in the case of hypochloruria) and injury to the renal tubules (in the case of hyperchloruria). There was no change in hippuric acid content of urine.

The results of antagonist muscle chronaximetry served as an indication of the functional state of the central nervous system with exposure to dioxane. The tests were made using an ISE-01 instrument by the conventional method.

As shown by the results of examining the 1st group of animals, the level of chronaxy on the 40th day of the experiment dropped from $0.341\pm0.003$ to $0.191\pm0.010$ ms for extensors and from $0.320\pm0.008$ to $0.201\pm0.006$ ms for flexors. Thereafter, as the experiment progressed, we observed an increase in time of reflex reaction of both groups of muscles, as compared to control

data and background findings. Thus, while the indices studied ranged from 0.34±0.008 to 0.335±0.003 ms for extensors and 0.325±0.008 to 0.317±0.003 ms for flexors in all groups of rats prior to exposure, by the end of the exposure period they constituted 0.370±0.004 and 0.370±0.005 ms, respectively (in both instances, $P<0.001$). Impaired correlations in different muscle groups were observed as a result of the changes in motor chronaxy.

The changes in chronaxy in the 2d group of rats were less marked and occurred toward the end of the experiment. However, they also led to impairment of proper correlation.

Animals submitted to inhalation of dioxane in concentrations of 4.6 and 20.5 $mg/m^3$ were found to also have less endurance with regard to physical loads.

Thus, this study of toxicity of dioxane revealed that the agent does not have a topical effect on the skin but, being rapidly absorbed in blood, it elicits acute poisoning and irritates the mucosa of the eyes. $LC_{50}$ constitutes 46 $mg/l$ for white rats and 65 $mg/l$ for white mice.

In the case of prolonged and continuous inhalation of dioxane in a concentration of 0.5 $mg/m^3$ for 90 days, it elicits mild, reversible, threshold changes in liver and kidney function.

Concentrations of 4 and 20 $mg/m^3$ dioxane, in the case of 90-day inhalation, elicited more marked changes in functional state of the liver, kidneys, nervous system, as well as reactions to physical loads. These concentrations are appraised as being effective.

## BIBLIOGRAPHY


1. Pokrovskiy, A. A. "Manual Dealing With the Study of Diet and Health of the Public," Moscow, 1964.

2. Stepanova, N. G. LABOR. DELO [Laboratory Record], No 5, 1962, pp 49-53.

3. Mikheyeva, A. I., and Bogodarova, I. A. Ibid, No 7, 1969, p 41.

4. Shumskaya, N. I., and Karamzina, N. M. in: "Metody opredeleniya toksichnosti i opasnosti khimicheskikh veshchestv" [Methods of Determining Toxicity and Hazard of Chemicals], Moscow, 1970, pp 142-168.

5. Tonhazy, N.; White, N.; and Umbreit, W. ARCH. BIOCHEM., Vol 28, 1950, pp 36-42.

6. Tursky, L., and Hruzik, J. BRATISL. LEK. LISTY, Vol 40, 1960, p 7.

UDC: 574.685:628.356.4

# HYGIENIC APPRAISAL OF WATER REGENERATED FROM DIVERSE FLUID-CONTAINING WASTE

Moscow KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA in Russian
No 6, 1977 pp 57-63

[Article by Z. P. Pak, Yu. S. Koloskova and S. V. Chizhov, submitted 16 Jun 75]

[Text] Implementation of the programs of biomedical research onboard the Salyut-3 and Salyut-4 orbital space stations made it possible to prove, for the first time, that it is possible to supply man with drinking water during space flights, which is obtained by recycling it from the condensate of atmospheric moisture in the spacecraft cabin.

The question of physiological acceptability of water regenerated from diverse liquid-containing products for the human body is gaining in importance, since the supply of ordinary potable water onboard space craft will be reduced.

Water obtained from diverse fluid-containing products, including human urine, condensate of atmospheric moisture in airtight areas, biological and technical waste is beginning to take a prominent place in providing potable water in space objects.

Along with the traditional methods of comprehensive hygienic appraisal (physicochemical, organoleptic and toxicological studies), hygienists are trying to expand the spectrum of methodological procedures in order to obtain fuller and more objective information about the physiological quality of regenerated water. In this respect, much attention is devoted to the study of indices of fluid and electrolyte metabolism in the human body [1-6].

Obviously, the method of balance-related characteristics is of first and foremost importance to studies of physiological acceptability of regenerated water for the human body. Experiments in sealed chambers should be considered the most convenient model for the study of fluid balance.

In this work, we submit the results of a comprehensive hygienic evaluation of diverse specimens of regenerated water, according to the findings of

physicochemical and organoleptic studies, as well as investigation of physiological effects of this factor on fluid and mineral metabolism in man.

Methods

The results are summarized of 5 experiments involving 25 healthy males ranging in age from 20 to 36 years; 2 of the experiments were conducted in a 5 $m^3$ sealed chamber under ordinary hospital conditions.

In all of the experiments, the subjects were on a diet consisting of natural products amount to a mean of 2800-3000 kcal/day. The experiments lasted 10 to 30 days.

Water regenerated from diverse liquid-containing waste (atmospheric moisture condensate in the chamber, human urine, technical fluids and biological waste) by catalytic-oxidative and sorption methods, to which salts were added and which was decontaminated with 0.1 mg/$\ell$ ionic silver, was used for drinking and cooking. A control group of subjects was supplied with water from the Moscow water supply system. There were no restrictions imposed on water intake, but an accurate record thereof was kept. For estimation of fluid balance, determination was made of daily intake of fluid in the form of water and food, diuresis, condensate of atmospheric moisture (CAM) in the sealed cabin and fecal liquid.

We studied the mineral balance of sodium, potassium, calcium, chlorides, sulfur and phosphorus in the subjects.

Flame photometry on a Zeiss-3 photometer was used for estimation of sodium, potassium and calcium in water, food, urine, feces and sweat; mercurometric titration was used for chloride ion, the Fiske-Subarrow colorimetric method was used for phosphorus, and photocolimetry with a barium-gelatin reagent was used for sulfur (converted to $SO_4$).

The water load test of Volhard was used to evaluate regulation of hydro-ion equilibrium and mineralocorticoid function of the adrenals, and we also assayed the principal minerals and aldosterone (by the method of Vel'tishcheva-Boykova) in blood and urine.

Studies of output of fluid and electrolytes under these conditions, combined with balance studies enabled us to gain an idea how mechanisms involved in fluid-electrolyte homeostasis function and, first of all, to evaluate the condition of the most important of these mechanisms, the renal one, under conditions of intake of water regenerated by various physicochemical methods.

Results and Discussion

Hygienic studies of regenerated water obtained from diverse fluid-containing waste revealed that, after artificial mineralization, the regenerated water is close to moderately mineralized potable water with regard to salt levels

(300 mg/ℓ); it has no extraneous flavor or odor and has high bacteriological indices (Table 1).

Table 1. Hygienic indices of quality of various specimens of regenerated water

| Indices | Water regenerated from: condensate in chamber | human urine | technic. fluids | biological waste |
|---|---|---|---|---|
| pH | 7,1 | 7,2 | 7,1 | 7,2 |
| Transparency, cm | 30 | 30 | 30 | 30 |
| Color, degrees | 5 | 5 | 5 | 5 |
| Odor } rating points | 0 | 0 | 0 | 0 |
| Taste } rating points | 0 | 0 | 0 | 0 |
| Overall hardness, mg-eq/ℓ | 2,7 | 2,87 | 2,9 | 3,1 |
| Calcium | 52,4 | 54,1 | 56,2 | 57,4 |
| Magnesium | 4,2 | 4,6 | 4,8 | 3,6 |
| Potassium | 35,0 | 28,4 | 32,5 | 28,2 |
| Sodium | 65,0 | 72,4 | 58,3 | 70,0 |
| Sulfates mg/ℓ | 34,2 | 34,0 | 36,0 | 40,0 |
| Chlorides mg/ℓ | 98,4 | 96,2 | 101,4 | 102,0 |
| Ammonia nitrogen | 0,2 | 0,3 | 0,15 | 0,2 |
| Nitrite nitrogen | 0,008 | 0,012 | 0,015 | 0,013 |
| Nitrate nitrogen | 0,2 | 0,25 | 0,02 | 0,25 |
| Permanganate oxidizability " | 1,5 | 2,0 | 1,2 | 1,3 |
| Bichromate " | mg $O_2$/ℓ 6,0 | 8,0 | 4,0 | 6,0 |
| Coli titer | 500 | 500 | 500 | 500 |
| Overall microbial number | 0 | 0 | 0 | 0 |

The main organic substances inherent in the sources of regenerated water were not demonstrable in the latter. Thus, in water regenerated from CAM of the sealed chamber, chromatography failed to demonstrate alcohols, aldehydes, ketones, fatty or organic acids and other components. Water regenerated from human urine did not contain organic nitrogen-containing substances (urea, uric acid, creatinine) inherent in urine; the ammonia level was in the range of 0.2-0.5 mg/ℓ. Water derived from various liquid-containing waste products did not differ basically in physicochemical indices with regard to different sources.

It is known that the physicochemical indices of water are closely related to its flavor. The gustatory features of water are physiologically important to meeting the body's liquid requirements and have a reflex effect on digestive function. Poor organoleptic indices of water, difference in odorimetric data and gustatory properties, as compared to the stereotype sample, may elicit an adverse reaction to water intake, to the extent of refusing to drink it. For this reason, preventive medicine attributes particular importance to high organoleptic properties of drinking water.

Table 2. Mean 24-h indices of external fluid metabolism of subjects who ingested water regenerated from diverse fluid-containing waste

| Duration of exper. days | Sources of water | Number of subjects | Fluid intake, g/day | | | Fluid output, g/day | | | | $\frac{U}{W} \cdot 100\%$ |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | water in food and beverages | metabol. fluid | total | in urine | in feces | perspiration moisture & sweat | total | |
| 14 | Cabin CAM | 8 | 1780,0± 100,0 | 314,0 | 2094,0 | 960,0 | 73,0 | 1000,0 | 2033,0 | 53,9 |
| 10 | Urine | 10 | 1850,0± 90,0 | 364,0 | 2214,0 | 986,0 | 86,0 | 1000,0 | 2072,0 | 53,3 |
| 10 | Technical fluids | 5 | 1950,0± 40,0 | 364,0 | 2314,0 | 1061,0 | 79,0 | 1100,0 | 2240,0 | 54,0 |
| 30 | Biological waste | 2 | 2302,0± 110,0 | 300,0 | 2602,0 | 1207,0 | 95,0 | 1200,0 | 2502,0 | 52,0 |
| *M±m* | | | 1972,0± 85,0 | 335,0± 12,0 | 2306,0± 97,0 | 1053,0± 47,0 | 83,2± 5,0 | 1075,0± 50,0 | 2211,0± 90,0 | 53,3± 0,48 |
| 10 | Control | 6 | 1945,0± 80,0 | 364,0 | 2302,0 | 1041,0 | 61,0 | 1265,0 | 2367,0 | 53,5 |

The high qualities of regenerated water have been confirmed, not only by the method of team appraisal, but organoleptic qualities rated by subjects who were questioned and whose answers were recorded in logs.

According to the subjects, the regenerated water tasted the same as ordinary drinking water and quenched their thirst well. Retention of normal liquid intake could be evaluated on the basis of the dynamics of daily intake of water and diuresis in the experimental and control groups during the experimental period (Table 2).

Observation of regimen of water intake revealed that the subjects in the experimental and control groups drank water primarily at mealtime, with breakfast, lunch and dinner, and very rarely were they thirsty between meals. When the subjects were questioned as to well-being and subjective sensations when drinking regenerated water, they failed to report any adverse reactions.

Food and beverages (tea, coffee, soup) prepared with regenerated water had the usual consistency and flavor. Throughout the experimental period, we failed to detect any deviations referable to peristaltic and secretory activity of the gastrointestinal tract.

Observation of dynamics of the subjects' daily intake of regenerated water and output in urine, feces, sweat and perspiration moisture revealed a normal course of external fluid metabolism in the experimental period, and it corresponded to the mean physiological norm (see Table 2).

The ratio of amount of excreted urine to total input of fluid (U/W) during the period that the subjects consumed various types of regenerated water

was in the range of 50-60% and did not differ from the analogous coefficient in the control group (see Table 2). These data were indicative of a normal correlation between various routes of fluid elmination and preservation of fluid equilibrium in the subjects' organism [7, 8]. The results of estimation of fluid content of blood were indicative of the lack of appreciable changes in hydration status of the organism; there were virtually no fluctuations of this index in experimental and control groups (86.3±3.3% in the control; 83.6±1.13, 84.0±0.73 and 82.9±0.8% in the experimental group).

The study of mineral balance in the organism of 10 subjects who ingested water regenerated from human urine by the catalytic-oxidative method for 14 days (Table 3) revealed that the mean levels of excretion in urine of sodium, calcium, potassium, chlorides, phosphorus and sulfur were in the range of mean physiological fluctuations with total fluid intake of 2214 mℓ, including the liquid in food, beverages and plain water. Under these conditions, the percentage of excretion of the main minerals in urine corresponded to the conventional norm. Thus, the 24-h mean constituted about 90% sodium, about 99% chlorides and up to 80% of ingested potassium eliminated in urine.

Table 3. Mean 24-h data on mineral metabolism of subjects who ingested water regenerated from human urine (M±m; mg-eq/day)

| Principal minerals | Input | | | Output | | | |
|---|---|---|---|---|---|---|---|
| | food | water | total | urine | feces | sweat | total |
| Na | 146±11 | 3,0±0,4 | 149,0 | 145,0±7 | 1,2±0,5 | 9,0±4,0 | 155,2 |
| K | 32,1±9 | 1,6±0,2 | 33,7 | 26,0±3 | 6,3±0,4 | 5,0±0,6 | 37,3 |
| Ca | 27,5±5,0 | 5,4±2,0 | 32,9 | 12,7±1,2 | 13,5±1,0 | 5,0±0,5 | 28,7 |
| Cl | 150,0±15 | 3,5±0,4 | 153,5 | 149,0±7,6 | 1,5±0,1 | 8,0±3,0 | 156,5 |
| P | 75,0±9 | Следы | 75,0 | 72,8±3,9 | 1,4±2,0 | 1,5±0,3 | 75,7 |
| S ($SO_4$) | 67,0±7 | 0,7 | 67,7 | 64,5±6,5 | — | — | |

The obtained data are indicative of absence of disturbances referable to fluid-mineral metabolism in subjects who used regenerated water from human urine, which had undergone the stage of standardization of quality, for drinking purposes and cooking.

A study of aldosterone content of urine during the experimental period confirmed the absence of disturbances referable to adrenal mineralocorticoid function, which constituted 9.18+1.3 μg/day; a similar conclusion could be made on the basis of calculation of the Na/K coefficient of urine, which was in the range of 0.18±0.023.

The study of mineral levels in blood serum of subjects at the time they used regenerated water for drinking and cooking purposes was supplemented by the data on the course of mineral metabolism and indicated that there were no disturbances of hydroion homeostasis (Table 4).

The results of water-load tests are very important in gaining understanding of the functional state of mechanisms of regulation of excretion of fluid

and salts, as related to the effects on the human body of regenerated drinking water.

Table 4. Levels of principal minerals in blood serum of subjects ingesting various samples of regenerated water (M±m, mg-eq/100 mℓ)

| Sources of water | Na | K | Ca | Cl |
|---|---|---|---|---|
| Control | 14,1±0,34 | 0,55±0,01 | 0,5±0,05 | 9,6±0,15 |
| Urine | 14,6±0,29 | 0,46±0,02 | 0,5±0,01 | 9,9±0,14 |
| CAM | 14,8±0,4 | 0,46±0,01 | 0,46±0,01 | 10,02±0,16 |
| Technical fluids | 14,5±0,2 | 0,42±0,03 | 0,45±0,09 | 10,1±0,12 |

It is known that the water-load test is of great diagnostic value as a complex method of evaluating renal function and humoral systems that regulate fluid-electrolyte homeostasis of the organism.

The results of determination of excretion of fluid and main, osmotically active substances (sodium and potassium), and correlation between elimination thereof during the water-load test in subjects given various specimens of regenerated water are illustrated in Figures 1 and 2.

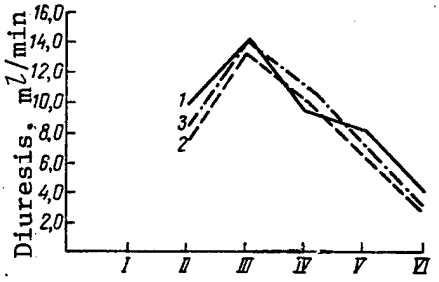



Figure 1.
Renal excretion of fluid during the water-load test (mean data).
X-axis, 30-min test intervals;
y-axis, diuresis, mℓ/min.
Here and in Figure 2:
1) water regenerated from urine
2) from CAM
3) from Moscow water system

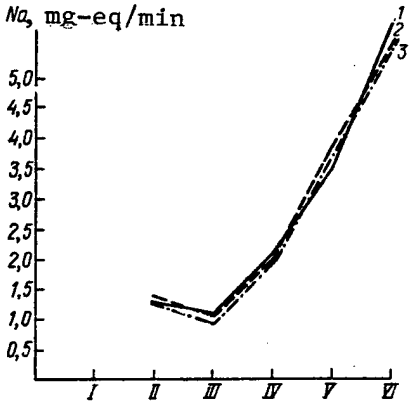



Figure 2.
Renal excretion of sodium during the water-load test.
X-axis, 30-min test intervals;
y-axis, sodium content, mg-eq/min

The main physiological effect was manifested in the first stage of the water-load test, after 30-120 min: termination of facultative resorption

of fluid and elimination of a large amount of minimally concentrated urine, diuresis of up to 13.3, 13.9, 14.0 ml/min during the maximum period (third batch)(see Figure 1). The figures characterizing the rate of elimination of the main electrolytes of sodium, potassium and their behavior, against the background of fluid diuresis, are illustrated in Figure 2.

There was significant decrease in excretion of sodium and potassium, in the presence of maximal fluid diuresis (2d-3d specimens): down to 1.3, 1.36 and 1.2 mg-eq/min for sodium, 0.12, 0.2 and 0.17 mg-eq/min for potassium.

Diuresis and concentration of osmotically active substances in the subjects' urine reached initial values by the end of the 4th hour of the water test (see Figures 1 and 2). Thus, the functional water test of Volhard, conducted on the subjects during the period of intake of regenerated water, enabled us to demonstrate the classical type of hourly diuresis, with maximum elevation in the 1st and 2d hours and significant decrease at this time in concentration of osmotically active substances, potassium and sodium.

These data were indicative of adequate stability of regulatory processes that prevented impairment of water-electrolyte equilibrium of the organism. These complex physiological and hygienic studies enabled us to evaluate the physiological acceptability of regenerated water for the human body.

Using the balance method, we were able to analyze the condition of systems that regulate fluid-electrolyte metabolism and to demonstrate that water regenerated from various fluid-containing products has no specific effect whatsoever on fluid-electrolyte equilibrium in the human body.

The data referable to complex physiological and hygienic appraisal of regenerated water are consistent with the results of previous toxicological experiments with hydrobiological objects and warm-blooded animals, as well as findings with regard to clinicophysiological indices and fluid-electrolyte balance in subjects participating in a year-long experiment [9].

The experimental data pertaining to dynamics of excretion of fluid and minerals warrant the conclusion that regenerated water, which has undergone a stage of standardization of quality, is physiologically satisfactory potable water, elicing an adequate reaction of the organism to intake thereof, as inherent in ordinary drinking water.

## BIBLIOGRAPHY

1. Nikolayeva, T. A.; Bokina, A. I.; et al. GIG. I SAN. [Hygiene and Sanitation], NO 11, 1970, pp 11-14.

2. Rozval, K. S. Ibid, No 8, 1972, pp 21-25.

3. Rakhmanin, Yu. A.; Plugin, V. P.; and Rozhkov, G. I. in: "Opresneniye solenykh vod i ispol'zovaniye ikh v vodosnabzenii" [Desalinization of Salt Water and Use Thereof in Water Supply Systems], Moscow, 1972, pp 180-186.

4. Sidorenko, G. I. GIG. I SAN., No 2, 1971, pp 16-20.

5. El'piner, L. I.; Bokina, A. I.; et al. Ibid, No 6, 1969, pp 22-26.

6. El'piner, L. I. KOSMICHESKAYA BIOL. [Space Biology], No 6, 1971, pp 73-77.

7. Zaretskiy, I. I. "Clinical Physiology and Renal Function Test Methods," Moscow, 1963.

8. Mohlman, N. T.; Katchman, B. J.; and Slonim, A. R. AEROSPACE MED., Vol 39, 1968, pp 386-402.

9. Chijhov, S. V., and Adamovich, B. A. in: "International Astronautical Congress, 22d," Brussels, 1971, pp 277-279.

UDC: 574.682.001.57

# VARIANTS OF CONTROL FOR AN ECOSYSTEM THAT IS CLOSED WITH REGARD TO EXCHANGE OF GASES, WITH PERIODICALLY FUNCTIONING AUTOTROPHIC COMPONENT

Moscow KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA in Russian
No 6, 1977 pp 63-68

[Article by V. G. Shabel'nikov, submitted 29 Jan 75]

[Text] This work is a continuation of a study of the possibility of controlling an ecosystem, closed with respect to gas exchange, by means of periodic function of an autotrophic component. The objective of such control is to maintain a stable atmosphere in the ecosystem, the composition of which changes periodically but always remains within the range tolerated by man [1, 2].

## Formulation of the Problem of Ecosystem Control

Compliance with the three following correlations between component parameters and atmosphere of an ecosystem is a mandatory and sufficient prerequisite for stability of the atmosphere of an ecosystem [2]:

$$\begin{aligned} &\text{a)}\ y_{\min} = \frac{\varphi_y}{\lambda\mu} + \Gamma + \frac{(\varphi_y + \gamma_\tau\mu)(T-\tau)}{e^{\lambda\mu\tau} - 1}; \\ &\text{b)}\ \Delta y = (\varphi_y + \gamma_\tau\mu)(T-\tau); \\ &\text{c)}\ \varphi_y\left(\frac{1}{R_m} - \frac{1}{r}\right)T + \gamma_\tau\mu\left(\frac{1}{R_\tau} - \frac{1}{r}\right)(T-\tau) = 0; \end{aligned} \qquad (1)$$

In the above equations (1) and following text, $x$ and $y$ refer to the concentration of $O_2$ and $CO_2$ in the ecosystem atmosphere; $\Delta x$ and $\Delta y$ are the ranges of changes in $x$ and $y$ within the light period; $y_{\min}$ is the value of $y$ at the end of the mode with light; $\phi_y$ is the ratio of rate of output of $CO_2$ by "man" $V_{CO_2}$ to volume of atmosphere $W$; $\mu$ is the ratio of "chlorella" mass $m$ to $W$; $\lambda$ is the slope of the linear segment of the carbon dioxide curve of photosynthesis; $\Gamma$ is the carbon dioxide compensation point; $\gamma_T$ is the specific rate of $CO_2$ emission by "chlorella" in the dark; $T$, $\tau$ and $T-\tau$ refer to duration of the cycle, light and dark conditions; $r$ is the "chlorella" assimilation coefficient; $R_T$, $R_m$ are respiratory coefficients; $R_m$ for "man" and $R_T$ for "chlorella" in the dark.

Equations (1) signify that the $CO_2$ level in a stable atmosphere have constant values for all cycles, at the start ($y_{min} + \Delta y$) and end ($y_{min}$) of the light period, and that the complete change in $x$ within each cycle equals zero. The parameter $\Delta x$ is not overtly included in the stability conditions, but it can be determined from equations 1b and 1c. As was shown in [2], the $\Delta x/\Delta y$ ratio differs little from one, and for this reason, even with maximum value of $\Delta y$ the fluctuations of $x$ will not be of physiological significance and, consequently, they can be disregarded in controlling the ecosystem.

Ecosystem control consists of providing for the values of ecosystem parameters and time of operation of the autotrophic component that would comply with equations (1), with permissible values of $y_{min}$ and $\Delta y$. In addition, the initial composition of the atmosphere should be the one that is normal for man.

For equation 1c to be possible, the type of autotrophic culture used as "chlorella" must be such that the difference between $R_m$ and $r$ and the difference between $R_T$ and $r$ would have opposite signs [2]. Values of $y_{min}$ and $\Delta y$ the sum of which does not exceed $y^*$ are permissible, as this is the maximum concentration of $CO_2$ tolerated by "man."

The above restrictions can be complied with over a wide range of values of ecosystem parameters, so that it is possible to select modes of ecosystem control that are optimal in some sense or other. To compare the different variants of control, let us express equation (1) in a dimensionless form, using the following "analogs" [?--similarity numbers]:

$$E = \frac{\lambda \Delta y}{\gamma_T}; \quad S = \frac{\lambda (y_{min} - \Gamma)}{\gamma_T}; \quad K = \frac{\varphi_u}{\mu \gamma_T}; \quad L = \frac{1 - \frac{r}{R_T}}{\frac{r}{R_m} - 1}; \quad N = \frac{\gamma_T}{\lambda \varphi_y}. \quad (2)$$

Substituting the variables according to formula (2) in equations (1), we obtain:

$$\text{a) } S - K - \frac{E}{\exp\left\{\frac{E}{K}\frac{L-K}{1+K}\right\} - 1} = 0;$$

$$\text{b) } \frac{\tau}{N} = E\frac{L-K}{K+1}; \quad (3)$$

$$\text{c) } \frac{T-\tau}{N} = \frac{EK}{K+1};$$

The "analogs" have the following meaning: $S$ is the lowest "rate of photosynthesis," which equals the ratio of lowest "visible" productivity

of "chlorella" photosynthesis to intensity of its respiration in the dark, as measured according to $CO_2$; $E$ is the range of changes in "rate of photosynthesis" during the light period; $K$ is the "useful load" per unit biomass of "chlorella"; $L$ is the "degree of concordance" of $r$ and $R_m$ of the system, which increases to infinity as $r$ draws closer to $R_m$ and decreases to zero as $r$ draws closer to $R_T$; $N$ is the time constant that determines the actual duration of the cycle.

Ecosystems, in which all of the above-mentioned five numbers are the same, are similar, since they have the same atmosphere composition at any given time. The parameters characterizing the size of the ecosystem, i.e., $m$, $V_{CO_2}$ and $W$, are contained only in $K$ and $N$; for this reason the condition for similarity of ecosystems differing only in dimensions consists of equality of these numbers. Ecosystems can also be similar when they have different values of $r$, $R_T$ and $R_m$, but the same $L$.

Results of Calculations

According to the above-imposed restrictions, $0<L<\infty$, $E+S<\frac{\lambda(y^*-\Gamma)}{\gamma_T}$, by virtue of (3a) $S>0$, since $E$, $K$ and $N$ are positive in the sense of parameters they include. With all permissible values of $L$, $S$ and $E$, system (3) has a single solution, which is consistent with the fact established in [2] that there is only one stable composition of the atmosphere. The solutions for system (3) were obtained numerically on a computer for various permissible values of $L$, $E$ and $S$, which satisfy the condition $E+S\leqslant S^*\approx 15$. This estimate of $S^*$ is consistent with the hypothesis that respiration in the dark constitutes about 6-7% of the rate of photosynthesis with $CO_2$ saturation, while the maximum tolerable concentration of $CO_2$, $y^*$, coincides with the point on the $y$ axis, in which the $CO_2$ curve of photosynthesis emerges on a plateau. Figures 1-3 illustrate the values of $K$, $\frac{\tau}{N}$ and $\frac{T-\tau}{N}$ obtained in solving system (3) in the form of explicit functions of $L$, which are parametrically dependent on $E$ and $S$.

$K$ as function of $L$, with all values of $E$ and $S$, is expressed by convex curves, which go out on a plateau, $K = S$, as $S$ increases (see Figure 1). The lower the value of $S$ and the higher that of $E$, the faster the plateau is reached. All of the $K(L)$ curves originate from the beginning of the coordinates, where their tangents have a slope that equals $E/(E+\ln(1+E/S))$.

The duration of the light mode increases almost linearly with increase in (Figure 2). The nonlinearity of curves $\tau(L)$ is noticeable only near the start of the coordinates. The duration of the dark mode also increases with increase in $L$, but it soon reaches the maximum level, which is unrelated to $L$. With $L\to 0$, $\tau$ and $T-\tau$ also strive toward zero, and the ratio $\frac{\tau}{T-\tau}$ tends toward the limit of $\frac{1}{E}\ln\left(1+\frac{E}{S}\right)$. An increase in $E$ leads to appreciable increase of both $\tau$ and $T-\tau$. Growth of $S$ leads to some increase of $T-\tau$ and significant decrease of $\tau$.

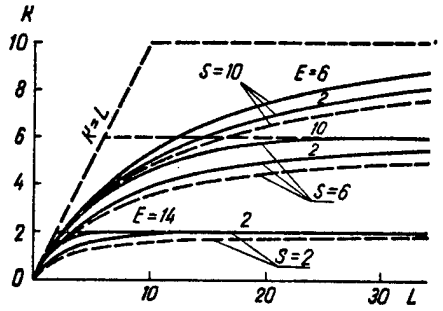



Figure 1. Functions $K(L)$ with different values of $E$ and $S$.

$K$) "useful load" per unit "chlorella" mass, determined by $CO_2$ output by "man"

$L$) concordance of respiratory coefficient of "man" and photosynthetic coefficient of "chlorella"

$S,E$) lowest value and range of changes within period of light in "rate of photosynthesis," which is proportionate to the $CO_2$ concentration in the atmosphere.

The dash lines indicate the boundaries of the sets of $K(L)$ curves at three levels of $S$ and any $E$ from 0 (bottom hyperbolas) to $\infty$ (top interrupted lines); the solid lines refer to functions $K(L)$ with $E$ = 2 and at maximum level of $E$ that is tolerated by "man" at a given $S$.

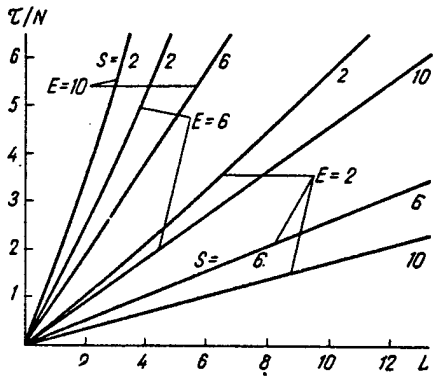



Figure 2.
Duration of light mode τ normed to a "time constant" $N$, as function of $L$ with different values of $E$ and $S$.

$L,E,S$) the same as in Figure 1.
The "time constant" is proportionate to the volume of ecosystem atmosphere.

Figure 4 illustrates the distribution of time in the light and dark modes.

Discussion

The "payload" and mean rate of assimilation of $CO_2$: The only dimensionless variable that depends on the mass of "chlorella" is "payload" $K$. If the "chlorella" functioned only in the light, the "payload" would equal the specific rate of $CO_2$ assimilation by "chlorella" normed for $\gamma_T$, i.e., the "rate of photosynthesis." With alternation of light and dark modes, the mean rate of $CO_2$ assimilation can be characterized by the

mean "rate of photosynthesis" $I = \frac{\lambda(\bar{y}-\Gamma)}{\gamma T}$, where $\bar{y}$ is the mean value of $y$ over the light period. Writing down the equation for balance of $CO_2$ in the atmosphere for the cycle, we shall obtain:

$$I = K + (K+1)\frac{T-\tau}{\tau} = K\left(1 + \frac{K+1}{L-K}\right). \qquad (4)$$

Thus, in the case of alternation of modes, $I$ is always greater than $K$; however, as in the case of continuous light, with the specified type of components, the choice of "payload" sets the mean intensity of photosynthesis, as well as the distribution of time referable to the modes of light and darkness, as can be seen from (4). Variations of the other controlled parameters, with a constant $K$, could alter only the form of fluctuations of "rate of photosynthesis" in relation to the constant mean level and overall duration of the cycle $T$. With increase in $L$, $I \to S$, just as is the case for $K$. Equality $K = S$ is established with relatively low values of $L$, for which the mean "rate of photosynthesis" is still appreciably higher than the minimum; however, it no longer depends on the range of fluctuations of $E$. This equality means that, within the period of light, there is time for the constant level of $CO_2$ to become established in the system, as it occurs in the case of continuous exposure of "chlorella" to light.

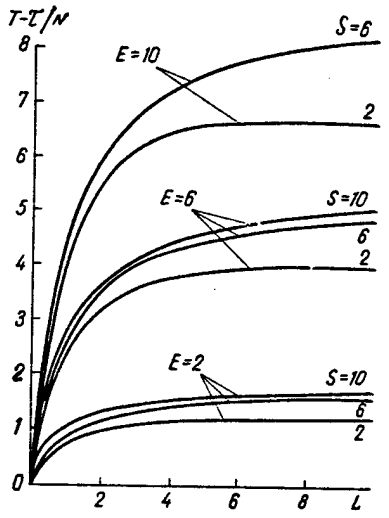



Figure 3.
Duration of dark phase $T-\tau$ normed to $N$ with different values of $L$, $S$, $E$. Designations are the same as in Figures 1 and 2. The height of the curve plateau is:

$$\frac{ES}{1+S}$$

Parameter $\eta$ serves as a convenient criterion of variants of ecosystem control; it equals the ratio of $CO_2$ output per cycle by "man" to the amount of $CO_2$ absorbed in the same time by "chlorella," i.e., it is the efficiency of "chlorella" as a $CO_2$ absorber. Using equation (4), we obtain:

$$\eta = \frac{L}{1+L}. \qquad (5)$$

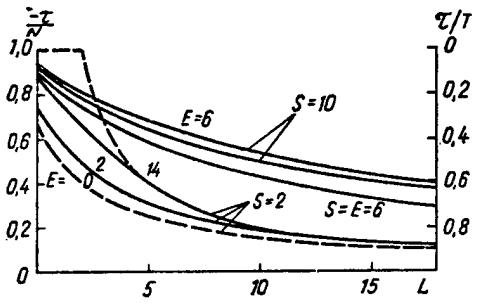



Figure 4.
Distribution of time in the light and dark with different values of $L$, $E$, $S$. Designations are the same as in Figure 1. On the left, the share of darkness in the cycle is plotted vertically; on the right, the share of light. The dash lines refer to boundaries of the family of "curves of distribution of time" with $S = 2$ and any value of $E$ from 0 to $\infty$.

The magnitude of $\eta$ depends only on $L$. This means that, with the specified type of components, control of their size and duration of modes has a substantial effect on composition of the stable atmosphere, but cannot alter the efficiency of the process of utilization of $CO_2$ excreted by "man," which increases with increase in $L$ and reaches one for systems with ideally coordinated $r$ and $R_m$.

Although the efficiency of "chlorella" is entirely determined by the nature thereof, one can apparently increase the economy of the system by reducing the "chlorella" mass, increasing $S$ for this purpose and, consequently, also increasing the "payload" $K$. An increase in "rate of photosynthesis" will be associated with decrease of $\tau$ (see Figure 1) and $\tau/T$ (see Figure 4), which will lead to reduction of total time of exposure of "chlorella" to light and diminish expenditure of luminous energy. Thus, the higher the "payload," the more "economical" the ecosystem. However, the increase in "payload" should be associated with reduction in duration of the cycle and in "chlorella" mass, which lowers the stability of the ecosystem atmosphere, since there is a decrease of the "damping coefficient," $h = \lambda\mu\tau$ [2].

Thus, the more economical the ecosystem, the more susceptible the composition of its atmosphere to random changes under the influence of perturbations that are inevitable in experimental ecosystems.

Control of ecosystem in the presence of random perturbations: For experimental ecosystems subject to the effect of random perturbations, changes in atmosphere composition are a random process, so that it is necessary to expand the previous definition of stability of atmosphere. It is expedient to consider the atmosphere of an ecosystem to be stable [stationary] if it complies only with the conditions stipulated in system (1). For ecosystems with random changes in parameters, equations (1b) and (1c) will retain their former appearance if $\phi_y$ in (1b) and $\gamma_T$ in (1b) and (1c) are replaced by their mean values for the darkness mode, and $\phi_y$ in (1c), by the mean value of this function for the entire cycle. The third condition for stability is the requirement of constant $y_{min}$, is specified, in the general case, by the functional of random perturbances and control operations [1]. This condition will retain the simple appearance of equation (1a) if the "damping coefficient" is so high that, by the end of the light period $y$ has time to reach the maximum constant level, which is unrelated to its

prior changes. In this case, the expression for $y_{min}$ is the equality $K = S$, which contains one random function, $\phi_y$. The condition of constant $y_{min}$ will be complied with if $\phi_y$ is constant at the end of the light period, i.e., if the period of light will end when man is asleep, and his gas exchange is constant.

Ecosystems that have high "damping coefficients" can be effected only with a low "payload." In such ecosystems, perturbations of atmosphere composition will be damped in much less time than the duration of the light period, so that control thereof does not basically differ from control of ecosystems with constant parameters, as discussed in this article. At the same time, control of experimental ecosystems with a high "payload" will require comprehensive knowledge about the behavior of the autotrophic component with alternate modes of function thereof.

Thus, ecosystems with a low "payload" are notable for high stability and simplicity of control, in "payment" of which is the greater mass of "chlorella" and greater expenditure of luminous energy, as compared to ecosystems with a high "payload," which, in turn, have low stability and require complex control.

BIBLIOGRAPHY

1. Shabel'nikov, V. G. KOSMICHESKAYA BIOL. [Space Biology], No 4, 1975, pp 10-13.

2. Idem, Ibid, No 6, pp 36-40.

3. Andronov, A. A.; Vitt, A. A.; and Khaykin, S. E. "Vibration Theory," Moscow, 1959.

UDC: 616-02:612.766.2]-092.9-085.214.22

# EFFECT OF SEDUXEN ON THE COURSE OF EXPERIMENTAL HYPOKINESIA

Moscow KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA in Russian
No 6, 1977 pp 68-73

[Article by L. V. Zorya, submitted 4 Apr 76]

[Text] The pathological changes occurring with hypokinesia are related in certain ways to the time and conditions under which it is induced. Thus, the functional changes observed for the first 2 weeks of limited movement are aggravated, in the opinion of some authors [1, 2], by the effects of emotional stress on the animal or human organism.

Of all the reactions of the organism to restricted mobility, impairment of oxygen balance plays a rather significant role, and it is also manifested the most distinctly in the first 16 days of hypokinesia [3].

On the basis of these considerations, we deemed it purposeful to use seduxen to remove emotional stress and, at the same time, normalize disturbances referable to oxygen balance. According to the literature, this drug has both tranquilizing-sedative [4, 5] and antihypoxic [6, 7] properties. Moreover, seduxen has now found broad application in clinical practice, particularly in the treatment of neurological and mental diseases [8-10].

## Methods

Hypokinesia was produced by putting animals in specially designed, metal mesh, individual cages that restricted their activity markedly. Special metal feeders with two compartments were used to give food to the experimental animals: one for liquids and the other for solid feed. The rats were kept on the vivarium diet (bread, milk, grain and water).

Five series of experiments were conducted on 227 white, mongrel rats of both sexes, with initial weight of 130-180 g. The first series of animals (65 rats) was not given seduxen. The 2d series (20 rats) was given seduxen per os, once a day in a dosage of 5 mg/kg. The drug was given to the 97 animals in the 3d series in the same dosage, but intraperitoneally. In the 4th series, 10 rats were given seduxen per os, 3 times a day, in a daily

dosage of 15 mg/kg. In the 5th series, the animals were given hypodermic injections of the drug twice a day; the daily dosage constituted 10 mg/kg. In the 2d-5th series, seduxen was given to the animals for 8 experimental days, without taking them out of their cages. The control consisted of 25 rats whose movements were unrestricted, and who were kept in the same room. The air temperature was 20-22°C.

We studied the animals' behavior, weight dynamics, mortality, EKG findings, succinate dehydrogenase activity [11], as well as processes of glycolysis and tissular respiration in the myocardium, liver, skeletal muscle of the thigh in a Warburg apparatus on the 4th, 8th and 16th days of hypokinesia.

The results were submitted to statistical processing, using the nonparametric criterion U of Wilcoxson-Mann-Whitney and criterion $t$ of Fisher-Student [13].

Results and Discussion

For the first 8 days, the animals in the 1st series were aggressive, they gnawed at the metal feeders and emitted squeaky sounds. The signs of excitement were somewhat less marked in rats given seduxen.

In the course of hypokinesia, we observed weight loss in all series of experiments.

Mortality, which was observed in the course of the experiment primarily in the first 2 weeks of restricted movement, did not diminish with administration of seduxen. Thus, 31% of the animals in the 1st series died within the first 16 days of hypokinesia, and 33% died in animals referable to the 2d-5th groups. The route of administration of the drug did not have an appreciable effect on dynamics of mortality. The Table illustrates changes in weight and mortality of animals in all series.

Pathoanatomical examination of carcasses of rats that died during the experiment revealed that, in most cases (87.5%), the cause of death was a change in the lungs--edema and bronchopneumonia ["focal pneumonia"]; death occurred due to effusions of blood in the adrenals in 9% of the cases, and in 3.5% the cause of death remained undetermined. Relevant data have been reported elsewhere [14], to the effect that, with 33.3% mortality rate among hypokinetic mice, one-third of the animals died of pneumonia (according to pathoanatomical findings) and the others, of unknown causes.

Studies of intensity of glycolysis in tissues of the liver, heart and skeletal muscle were conducted on animals in the 1st (who did not receive seduxen) and 3d series of animals (given the product intraperitoneally).

The level of glycolytic processes in femoral skeletal muscle of the animals in the 3d series increased sharply on the 4th experimental day, to $2.91\pm0.202$ $\mu l$ $CO_2$ per mg dry tissue in 30 min, versus $2.27\pm0.208$ $\mu l$ $CO_2$/mg dry tissue in 30 min in the control ($P_u<0.01$). Intensity of glycolysis

Dynamics of weight and mortality of animals in the course of hypokinesia

| Series | Experimental conditions | Number of rats | Day of experiment: deaths 1 | 2 | 3 | 4 | weight change, % | 5 | 6 | 7 | 8 | weight change, % | 9 | 10 | 11 | 12 | 13 | 14 | 15 | 16 | weight change, % |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| I | Saline, 0.2 ml | 65 | | | 2 | 1 | —9,7±0,65 | 3 | 2 | 5 | 2 | —17,0±1,27 | 3 | | | | | | 1 | 1 | —18,0±1,15 |
| II | Seduxen per os, 5 mg/kg/day | 20 | | | | | —9,2±1,01 | | | 1 | 1 | —17,5±0,75 | 2 | 1 | | 2 | | | | | —18,6±1,97 |
| III | Seduxen intraperitoneally, 5 mg/kg/day | 97 | | | 1 | 4 | —5,8±0,77 | 3 | 5 | 4 | 4 | —13,9±0,59 | 3 | 3 | 1 | 4 | | 1 | | 1 | —17,3±1,26 |
| IV | Seduxen per os, 15 mg/kg | 10 | | | | 1 | —8,4±1,34 | | 1 | | | —12,5±1,30 | | | | 1 | | | | | —17,8±2,06 |
| V | Seduxen hypodermically, 10 mg/kg/day | 10 | | | | | —6,2±1,20 | | 1 | 1 | | —12,6±1,60 | | | | | 1 | | | | —15,7±1,75 |
| Control | | 25 | | | | | +2,0±0,13 | | | | | +4,3±0,15 | | | | | | | | | +10,5±0,31 |

remained high thereafter (8th and 16th days of hypokinesia): 2.71±0.310 and 2.63±0.166 μl $CO_2$/mg dry tissue/30 min ($P>0.05$).

Opposite findings were made in the muscle tissue of the 1st series of animals that was not given seduxen. In these animals, the level of glycolytic processes consistently declined under the influence of hypokinesia, from 2.74±0.208 μl $CO_2$/mg dry tissue/30 min in the control to 2.05±0.148 μl $CO_2$/mg dry tissue/30 min on the 16th experimental day ($P<0.02$)(Figure 1c).

The obtained data, which were indicative of increased intensity of glycolytic processes in muscle tissue of animals given seduxen, served as the grounds for examining the other, main route of obtaining energy, tissular respiration. It was found that the level of tissular respiration in femoral skeletal muscle of animals in the 3d series dropped sharply on the 4th day of hypokinesia, to 1.44±0.074 μl $O_2$/mg dry tissue/30 min, versus 1.77±0.071 μl $O_2$/mg dry tissue/30 min in the control ($P_u = 0.05$). Decreased intensity of tissular respiration was also noted on subsequent days: it constituted 1.31±0.141 μl $O_2$/mg dry tissue/30 min on the 8th day ($P_u = 0.05$) and 1.50±0.230 μl $O_2$/mg dry tissue/30 min on the 16th day ($P_u<0.05$). There was less marked decrease in tissular respiration in animals that were not given seduxen (Figure 1d). The changes in intensity of utilization of oxygen by muscle tissue during hypokinesia were confirmed by histochemical studies. Thus, on transverse sections of femoral muscle of animals in the 3d series, we found significant decrease in activity of succinate dehydrogenase of the axial portion of some fibers and occasionally depletion of this region, with appearance of so-called peltate [shield-shaped] fibers (Figure 2). Analogous changes were seen in muscle tissue of

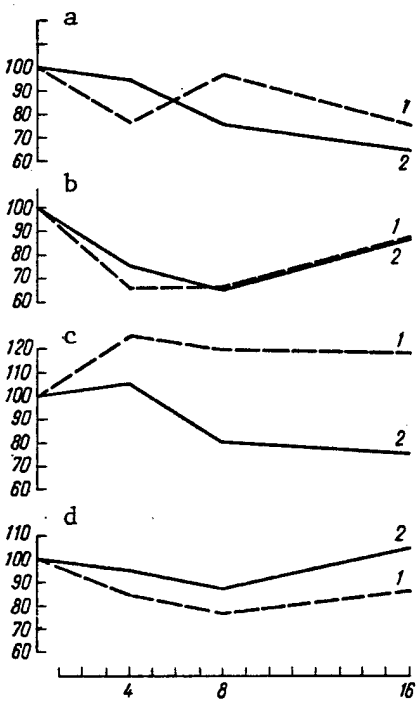



Figure 1.
Intensity of glycolysis and tissue respiration in rats given (1) and not given (2) seduxen during hypokinesia (%)
a) liver c) muscle
b) heart d) muscle

the 1st series of animals which was not given seduxen.

Changes in glycolysis in hepatic tissue of animals in the 1st and 3d series were identical in nature. Thus, in animals submitted to hypokinesia and not given seduxen, the level of glycolysis in the liver dropped from 2.58±0.302 μl $CO_2$/mg dry tissue/30 min in the control to 1.63±0.229 μl $CO_2$/mg dry tissue/30 min on the 16th experimental day ($P<0.05$). The same was observed when the animals were given seduxen, where the glycolysis level in the liver constituted 1.81±0.196 μl $CO_2$/mg dry tissue/30 min, versus 2.36±0.175 μl $CO_2$/mg dry tissue in 30 min in the control ($P_u<0.05$) (Figure 1a).

A study of glycolytic processes in the myocardium also revealed identical biochemical changes in animals given and not given seduxen. In both series, the intensity of glycolysis diminished, as compared to base data (Figure 1b).

We found substantial differences in myocardial function in the 1st and 3d series of animals. Marked bradycardia developed under the influence of seduxen in hypokinetic animals throughout the period of investigation. In the control group, the heart rate constituted 488±8.4/min; it constituted 431±9.2/min in rats given seduxen daily, in a dosage of 5 mg/kg, 40 min after administration of the drug, on the 8th day of hypokinesia ($P_u<0.01$). At this same time, tachycardia (510±8.0/min; $P_u<0.05$) was observed in animals not given seduxen.

During hypokinesia, qualitative EKG changes were observed along with quantitative ones. Thus, under the influence of restricted movement, there was an increase in amplitude of the T wave, which was often superimposed over the descending part of the QRS complex, forming an RST segment that is shifted from the isoelectric line. The T wave acquired a pin-like, pointed shape (Figure 3b). The changes arising in the myocardium were not removed by seduxen: signs of marked, diffuse changes in the myocardium were also seen on the EKG of animals in the 3d series (Figure 3c). Analogous changes, indicative of myocardial hypoxia in the 3d series of animals, were demonstrated histochemically. Diffuse scattering of formazan granules along the muscle fiber was observed in cardiac tissue.

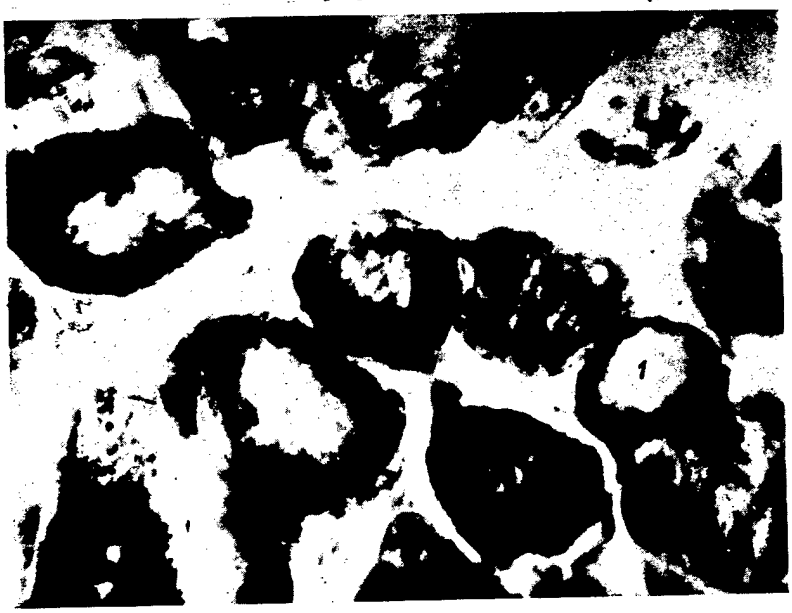



Figure 2. Disappearance of formazan granules from axis cylinder of muscle fiber (1) by the 8th day of hypokinesia in rats given seduxen. Nitroblue tetrazolium stain; objective 40× and ocular 10×

The results of the experiments are indicative of presence of emotional stress in the animals for the first 2 weeks of hypokinesia. However, it was not possible to completely prevent the effect of this "superposition," by giving seduxen in daily doses of 5-10 mg/kg. V. V. Zakusov [15] and S. D. D'yakova [16] demonstrated that, in the case of unrestricted animals, low doses of seduxen stimulate summation of impulses in the central nervous system. With prolonged administration of this drug, relative adrenal insufficiency develops [17], which is indicative of depletion of the hypophyseoadrenocortical system. All this attenuates development of adaptation mechanisms and accelerates depression of adrenal function, which is observed without this after 2-3 weeks of hypokinesia.

The increased glycolysis we observed in skeletal muscles is apparently attributable to the fact that the muscle-relaxant action of the drug under hypokinetic conditions is manifested more vividly and causes even greater limitation of movement. This, in turn, moves biochemical processes to a different level of regulation of obtaining energy, anaerobic glycolysis. Thus, intensification of glycolytic activity and decreased oxygen consumption in muscles are indicative of development of hypoxia under the influence of seduxen. Histochemical findings confirm the biochemical ones. Demonstration of "peltate" fibers in the study of succinate dehydrogenase activity is attributable to poorer conditions of nutrition of the axial part of the muscle fiber in the presence of generalized hypoxia [18].

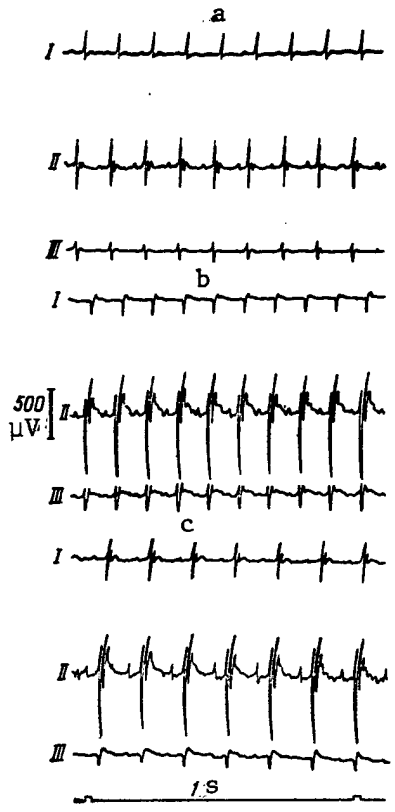



Figure 3.
EKG of rats given (c) and not given (b) seduxen, on the 8th day of hypokinesia. Recorded in three (I, II, III) standard leads.
a) control

V. Yu. Ostrovskiy et al. [6] report the opposite, antihypoxic action of seduxen given to patients 30-40 min prior to occlusion of the vena cava in open heart surgery, in a dosage of 0.5 mg/kg weight. An analogous effect was observed by the authors in experiments on mice, given 10 mg/kg of this drug. Evidently, under our experimental conditions, the combination of muscle-relaxant effect of seduxen and prolonged restriction of movement creates a rigid model of hypokinesia and enhances development of pathogenetic changes in the presence of this pathology. In this case, another mechanism of action of the drug may be involved.

The lack of beneficial changes in metabolism in the liver and myocardium under the influence of this product is also indicative of the undesirability of giving it in the case of restricted movement. The marked, diffuse changes in the myocardium (which we demonstrated on the EKG and histologically), which are not removed by seduxen, constitute more proof of this conclusion. This is apparently due to the fact that seduxen, even when movement is unrestricted, reduces the volumetric rate of coronary circulation and oxygen uptake by the heart [19]. These phenomena are accentuated in the case of restricted movement. Other authors have also observed slowing of heart rate, induced by seduxen, in the case of an unrestricted regimen [20].

Thus, in doses of 5-10 mg/kg/day, given via different routes to hypokinetic animals, seduxen has an adverse effect on several vital functions.

In the case of restricted movement, this product does not prevent development of hypoxia in the myocardium and skeletal muscles, i.e., it does not have antihypoxic properties. Seduxen does not normalize the biochemical changes that develop in the liver under the influence of severely restricted movement.

The results of these experiments cause us to question the desirability of using seduxen under conditions of restricted movement.

BIBLIOGRAPHY


1. Kovalenko, Ye. A.; Popkov, V. L.; Kondrat'yev, Yu. I.; et al. PAT. FIZIOL. [Pathological Physiology], No 6, 1970, pp 3-9.

2. Portugalov, V. V.; Il'ina-Kakuyeva, Ye. I.; and Artyukhina, T. V. in: "Eksperimental'noye issledovaniye gipokinezii, izmenennoy gazovoy sredy, uskoreniy peregruzok i drugikh faktorov" [Experimental Study of Hypokinesia, Altered Gas Environment, Accelerations, G-Forces and Other Factors], Moscow, 1968, pp 29-33.

3. Zorya, L. V. FIZIOL. ZH. [Physiological Journal], No 3, 1976, pp 414-417.

4. Magyar, I. VENGERSKAYA MED. [Hungarian Medicine], Vol 28, 1968, pp 57-68.

5. Sobor, A. VENGERSKAYA FARMAKOTER. [Hungarian Pharmacotherapy], Vol 1, No 4, 1969, pp 123-128.

6. Ostrovskiy, V. Yu.; Zil'bershteyn, M. S.; Yakubon, I. M.; et al. EKSPER. KHIR. [Experimental Surgery], No 5, 1974, pp 60-63.

7. Rudnev, M. I., and Degtyar', V. N. FARMAKOL. I TOKSIKOL. [Pharmacology and Toxicology], No 5, 1974, pp 552-553.

8. Aleksandrovskiy, Yu. A., and Prokudin, V. N. ZH. NEVROPATOL. I PSIKHIATR. [Journal of Neuropathology and Psychiatry], No 8, 1968, pp 1231-1237.

9. Varga, E. VENGERSKAYA MED., Vol 7, No 24, 1967, pp 37-41.

10. Dzhandzhgava, R. K. in: "Aktual'nyye voprosy klinicheskoy i eksperimental'noy farmakoterapii" [Pressing Problems of Clinical and Experimental Pharmacotherapy], Tbilisi, 1972, pp 133-134.

11. Sokolovskiy, V. V. "Histochemical Research in Toxicology," Leningrad, 1971.

12. Gubler, Ye. V., and Genkin, A. A. "Use of Nonparametrical Criteria of Statistics in Biomedical Research," Leningrad, 1973.

13. Belen'kiy, M. A. "Elements of Quantitative Evaluation of Pharmacological Effects," Leningrad, 1963.

14. Kravchuk, L. A., and Ovechkin, V. G. KOSMICHESKAYA BIOL. [Space Biology], No 3, 1968, pp 7-12.

15. Zakusov, V. V. FARMAKOL. I TOKSIKOL., No 1, 1971, pp 7-9.

16. D'yakova, S. D. in: "Stress i yego patogeneticheskiye mekhanizmy" [Stress and Its Pathogenetic Mechanisms], Kishinev, 1973, pp 144-145.

17. Robu, A. I., and Kuznetsova, T. S. FARMAKOL. I TOKSIKOL., No 2, 1972, pp 206-207.

18. Apatenko, A. K. ARKH. PAT. [Archives of Pathology], No 5, 1953, pp 37-41.

19. Chichkanov, G. G. FARMAKOL. I TOKSIKOL., No 4, 1975, pp 423-427.

20. Rozonov, Yu. B.; Chichkanov, G. G.; and Rayskiy, V. A. Ibid, No 2, 1973, pp 225-229.

BRIEF REPORTS

UDC: 613.693:629.785"Salyut-4"

# EFFECTS ON THE HUMAN BODY OF COMPLEX TESTS SIMULATING FLIGHT CONDITIONS ON THE SALYUT-4 SPACECRAFT

Moscow KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA in Russian
No 6, 1977 pp 74-75

[Article by V. P. Dzedzichek, N. Ye. Panferova and Ye. V. Kukolevskaya, submitted 27 Jan 76]

[Text] These studies were conducted in a spacecraft mockup. The living conditions were made as similar as possible to inflight conditions. There were two male participants in tests lasting 50 days, commander K and flight engineer B.

During the tests, the pulse rate of the subjects was counted every 3 h, arterial pressure was measured twice a day by the Korotkov method. A functional test, 40 squats/90 s, was performed every 2 days. The subjects performed physical exercises, the amount of which was adjusted to the inflight level.

We measured the volume of the foot in seated position and while standing for 1.5 min, before and after the tests, using a water plethysmometer. Determination was made of rate of propagation of the pulse wave in seven parts of the vascular system. We examined the capillaroscopic appearance of the eponychium of the 4th finger and first toe. We conducted a passive 20-min orthostatic test and a test on a bicylce ergometer with stepped up increase in load: 200, 800 and 1200 kg/min. The test periods and intervals between them constituted 5 min.

## Results and Discussion

The subjects' wellbeing was satisfactory during the tests. They adapted to the conditions by the 6th-10th day, which is consistent with the time of adaptation to flight conditions [1].

Starting on the 5th day for K and 25th day for B, we observed progressive loss of weight (Figure 1), although their caloric intake of food during the tests was consistent with energy expended (2840 kcal/day/person according to assimilated portion; 2610 kcal/day energy expenditure).

Mean daily pulse rate dropped to 45/min after the 9th-11th day, and some readings were 38-40/min. The pulse reverted to 56-58/min after the 47th-49th day (see Figure 1). A comparison of fluctuations of pulse rate to changes in oxygen and carbon dioxide levels in air failed to demonstrate any correlation between these parameters. K developed multiple, polytopic atrial and ventricular extrasystoles, most marked on the first 11 days (up to 2-5 extrasystoles per minute). The frequency thereof then diminished and became stabilized at 0-1 extrasystoles per minute. Evidently, a change in neural regulation of cardiac function during the tests, manifested by pulse slowing, was one of the causes of the extrasystoles.

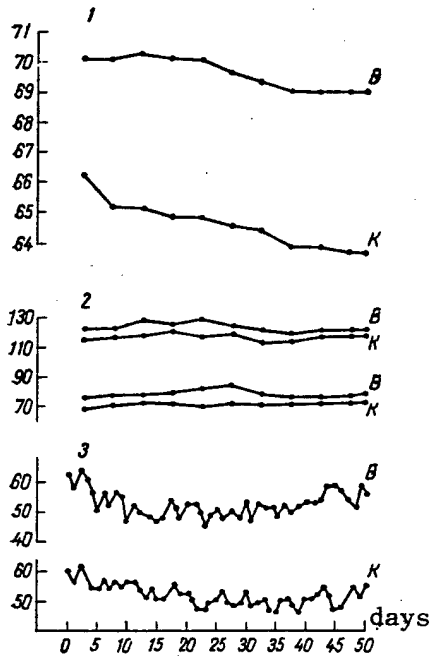



Figure 1.
Dynamics of body weight (1, in kg), arterial pressure (2, mm Hg; systolic, top curves; diastolic, bottom curves); mean daily pulse rate (3, beats per min) throughout the tests

Starting on the 10th day, both subjects presented diminished adaptation capabilities of the cardiovascular system to physical loads. This was manifested by more marked acceleration of pulse during the squatting test (in B) and slower recovery after the test.

Comparison of the data obtained before and after the tests at rest revealed a drop of systolic and pulse pressure, stroke (calculated by the method of Wetzler and Boger) and minute volumes of the heart.

After the tests, both subjects presented a faster rate of pulse wave propagation in the region between the femoral and posterior tibial arteries. The changes were in various directions in other parts of the circulatory system.

This suggests that there was an increase in tonus of arterial vessels of the legs after the tests. Concurrently, both subjects presented constriction of the capillary system of the lower limbs: shorter arterial and venous branches, less tortuosity of venous branches and fewer functional capillaries. The latter were markedly constricted. There were analogous, but less marked changes in the capillary system of the hand.

After the tests, B presented a decrease in volume of the leg (foot and two-thirds of the crus) by 140 mℓ, whereas in K it increased by 10 mℓ (Figure 2). The volume of the legs increased by 25 mℓ in both subjects while standing for 1.5 min, both before and after the tests. This is indicative

of a lack of appreciable changes in shifting of fluid to the lower limbs when changing to a vertical body position.

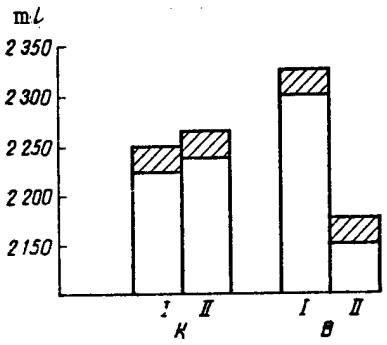



Figure 2.
Foot volumes before and after tests; striped segments, volume increment when standing for 1.5 min; white segments, volume of foot when sitting down
I) before tests II) after tests

After the tests, we demonstrated a decline of adaptational capabilities of the cardiovascular system in both subjects, with respect to the orthostatic test, there was a more significant increase in pulse rate (by 14-17/min) and lowering of systolic pressure (by 4-5 mm Hg).

The increase in tonus of arterial vessels and capillaries, and lack of changes in volume of the circulatory system of the legs when standing do not allow us to attribute the poorer adaptation to the orthostatic test to diminished tonus of vessels of the lower extremities. Among other causes of diminished adaptability of the cardiovascular system to the orthostatic test, apparently fatigue and asthenization of the subjects could be of greatest significance.

BIBLIOGRAPHY

1. Yeremin, A. V., and Kolosov, I. A. AVIATSIYA I KOSMONAVTIKA [Aviation and Cosmonautics], No 7, 1971, pp 6-7.

2. Dietlein, L. F. in: "Skylab Sciences Symposium," Houston, Vol 11, 1974, pp 369-388.

3. Michel, E. L., and Rummel, J. A. in: "Skylab 1/2, Preliminary Biomedical Report" (NASA), Houston, 1973, pp 303-308.

4. Butusov, A. A.; Yegorov, A. D.; Lyamin, V. R.; et al. in: "Aktual'nyye voprosy kosmicheskoy biologii i meditsiny" [Pressing Problems of Space Biology and Medicine], Moscow, Vyp 1, 1971, pp 36-38.

5. Smith, R. F., and Alleback, N. in: "Skylab 1/2, Preliminary Biomedical Report," Houston (NASA), 1973, pp 237-239.

6. Cherepakhin, M. A., and Pervushin, V. I. KOSMICHESKAYA BIOL. [Space Biology], No 6, 1970, pp 46-49.

7. Parin, V. V.; Bayevskiy, R. M.; and Gazenko, O. G. COR ET VASA, Vol 7, 1965, pp 165-183.

UDC: 629.78:611-018.46

# MORPHOLOGY OF BONE MARROW CELLS OF RATS ON THE KOSMOS-605 BIOSATELLITE

Moscow KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA in Russian
No 6, 1977 pp 75-78

[Article by V. N. Shvets and N. P. Krivenkova, submitted 20 May 75]

[Text] One of the distinctive features of weightlessness is the constant reduction of erythrocyte mass. Depression of medullary erythropoiesis may be the chief cause of this. Thus, in the opinion of P. A. Korzhuyev [1, 2], weightlessness may have some effect on vital function of bone marrow, depressing the normal process of hemopoiesis during space flights.

This work deals with a study of bone marrow morphology in rats who had spent 22 days in flight on the Kosmos-605 biosatellite and examined on the 2d and 27th day after landing.

## Methods

In order to study the size of the population of hemopoietic stem cells in rats that had been in orbital flight, we used the method of cultivation of bone marrow cells in the organism of lethally irradiated animals [3]. For this purpose, (CBA×C57Bl)$F_1$ mice and Wistar rats were exposed to $^{137}Cs$ γ-rays, in a dosage of 900 R delivered at the rate of 37 R/min. One day after irradiation, recipients were given intravenous injections of a suspension of bone marrow cells taken from rats on the 2d and 27th day after a 22-day flight onboard Kosmos-605.

On the 9th day after transplantation of bone marrow cells, we extracted the spleen from recipients, fixed it in Bouin liquid and counted macroscopically discernible cell colonies on its surface. To study bone marrow, we used femoral bones, from which we prepared serial sections 5-7 μm in thickness, after decalcification and imbedding in paraffin; the sections were then stained with hematoxylin-eosin and we counted the number of cells in mitosis (metaphase, anaphase, telophase).

Results and Discussion

Morphology of bone marrow: Microscopic examination of bone marrow from rats that had spent 22 days onboard the satellite, on the 2d day after the flight, failed to demonstrate appreciable changes in cell composition. At this time, the presence of many mature segmented neutrophils was a typical finding, and this could be indicative of faster maturation of juvenile forms of leukocytes. Among the mature neutrophils, we encountered groups of immature cells of the myeloblast and promyelocyte type, with rather large and full nuclei. The erythroid precursor is somewhat depressed and represented mainly by a few sparsely arranged groups of cells. There were fewer mature neutrophils in the bone marrow of control rats. The ratio of myeloid to erythroid class of cells was greater than one in both control and experimental animals, and it was higher in experimental rats. In spite of some depression of the erythroid class, mitotic activity of bone marrow cells of experimental rats did not differ from that of control animals (Table 1). Nor did we demonstrate differences in total number of nuclear bone marrow cells in experimental and control rats.

Table 1. Mitotic activity of rat bone marrow cells

| Index | After flight | | | |
|---|---|---|---|---|
| | 2d day | | 27th day | |
| | control | experiment | control | experiment |
| Number of rats examined | 4 | 4 | 6 | 4 |
| Mitotic index | 0,0026±0,0003 | 0,0033±0,0006 | 0,0023±0,0003 | 0,0032±0,0004 |

| Index | After ground-based experiment | | | |
|---|---|---|---|---|
| | 2d day | | 27th day | |
| | control | experiment | control | experiment |
| Number of rats examined | 5 | 4 | 5 | 6 |
| Mitotic index | 0,0027±0,0005 | 0,0027±0,0005 | 0,0026±0,0002 | 0,0023±0,0004 |

We also encountered structural deviations in some types of bone marrow cells of experimental rats. This applies, first of all, to megakaryocytes. They were represented by two forms of cells: 1) normal megakaryocytes with delicate structure of the nucleus and granular cytoplasm, and 2) cells with pyknotic nuclei and markedly eosinophilic, unstructured cytoplasm. The megakaryocytes referable to the 2d group were demonstrated in all of the experimental rats examined, and they constituted 10-20% of the megakaryocytes with normal structure. We failed to demonstrate the 2d type of megakaryocytes in the bone marrow of control animals.

Altered megakaryocytes were not encountered in rats on the 27th postflight day. At this time, the animals failed to demonstrate any appreciable changes

in quantitative proportion of myleoid and erythroid elements. We also failed to demonstrate changes in mitotic activity of bone marrow cells, as compared to control animals.

Table 2. Amount of colony-forming cells (CFU) in bone marrow of experimental rats demonstrated by cultivation of cells in the organism of irradiated mice

| Variant of experiment | Number of recip. | Number of colonies in recipient mouse spleen | Total number of cells in femur of donor rats ($\times 10^7$) | CFU per femur |
|---|---|---|---|---|
| 2d postflight day | 28 | 12,0±3,0 | 17,4±0,42 | 2100 |
| control | 21 | 16,8±4,0 | 16,2±0,3 | 2700 |
| 27th postflight day | 20 | 3,4±0,9 | 16,5±0,5 | 561 |
| control | 20 | 2,2±0,8 | 16,5±0,6 | 363 |

Note: Here and in Table 3, the animals were given $10^6$ cells.

The foregoing warrants the opinion that the above-described changes in morphology of bone marrow of experimental animals on the 2d postflight day are reversible. Experiments simulating space-flight conditions on the ground were not associated with a change in morphology of medullary cells. This also applies to the proliferative capacity of cells; mitotic activity was found to be the same in experimental and control animals (see Table 1).

Colony-forming properties of bone marrow cells of rats on the 2d and 27th days after flight on Kosmos-605 biosatellite: Tables 2 and 3 show that, after transplantation of rat bone marrow cells into mice (xenogenic combination) or rats (allogenic combination), macroscopically visible cell colonies are formed in the spleen of recipients exposed to a lethal dose of radiation. These experiments established that bone marrow cells of rats that had been on the space flight do not lose their colony-forming properties, and their proliferative activity remains at the same level as proliferation of colony-forming cells in control animals. Morphological analysis of cells of colonies formed in the spleen of irradiated rats and mice after transplantation of bone marrow cells from rats that had returned from the orbital flight revealed that there is no impairment of the capacity of stem cells for differentiation into cells of the erythroid and myeloid classes, and that it is comparable to the same properties of cells in intact animals.

In summary, it can be considered that only negligible changes, manifested by depression of erythroblasts and appearance of altered megakaryocytes, occur in the bone marrow cells of rats following a 22-day space flight. In spite of this, the total number of bone marrow cells remains stable, and their mitotic activity remains on the initial level. The colony-forming activity and capacity for differentiation of hemopoietic stem cells of bone marrow

Table 3. Number of colony-forming cells (CFU) in bone marrow of experimental rats demonstrated by the method of cultivation of bone marrow cells in the organism of irradiated Wistar rats

| Variant of experiment | Numb. of recipients | Number of colonies in recipient rat spleen | Total number of cells in femur of donor rats ($\times 10^7$) | CFU per femur |
|---|---|---|---|---|
| 2d postflight day | 25 | 44,0±10,0 | 17,4±0,42 | 7650 |
| control | 19 | 25,0±7,0 | 16,2±0,3 | 4100 |
| control irradiation | 12 | 0,9±0,6 | — | — |

cells of experimental animals also remained unchanged, as compared to the control. Normalization of cellular composition of bone marrow 27 days after the flight is indicative of the fact that the demonstrated changes in experimental rats are transient and, mainly, they do not affect the capabilities of stem cells responsible for the rpoduction of bone marrow cells. The demonstrated changes are referable primarily to elements of the mature or maturing classes of cells. It is believed that the changes in erythropoiesis and thrombocytopoiesis in the bone marrow of experimental rats could be due to one or several factors: weightlessness, hyperoxia, some restriction of motor activity and accelerations which occur when the spacecraft is landing on earth.

BIBLIOGRAPHY

1. Korzhuyev, P. A. ZH. OBSHCHEY BIOL. [Journal of General Biology], Vol 29, 1968, p 587.

2. Idem, "Evolution, Gravitation and Weightlessness," Moscow, 1971.

3. Till, I. E., and McCulloch, J. RADIAT. RES., Vol 14, 1961, p 213.

UDC: 612.12.018:612.453-06:629.78

# BLOOD PLASMA CORTICOSTERONE IN RATS AFTER FLIGHT IN KOSMOS-690 BIOSATELLITE

Moscow KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA in Russian
No 6, 1977 pp 78-79

[Article by N. F. Kalita and R. A. Tigranyan, submitted 13 Jul 76]

[Text] We are submitting here data from a study of corticosterone concentration in blood plasma of rats after completion of a 20.5-day space experiment in the Kosmos-690 biosatellite.

## Methods

We used the fluorimetric method of Ya. Popens et al. [1] to assay the concentration of corticosterone in rat blood plasma on a fluorescent spectrophotometer. Blood plasma was examined 1 and 26 days after the flight and at the same intervals after the "control-2" experiment. The data were compared to findings made on intact rats kept in the vivarium on a special diet. Each of the experimental groups had its own intact control.

## Results and Discussion

The concentration of corticosterone in blood plasma of rats exposed to 800 rad radiation, 1 and 26 days after the space flight, was the same as in intact animals. In rats exposed to a dosage of 220 rad, the corticosterone level 1 day after the flight was significantly higher than in intact animals, whereas it did not differ from indices in the control group on the 26th post-flight day (see Table).

In the "control-2" experiment, after irradiation in a dosage of 800 rad 1 day after completion thereof, there was also no difference in hormone levels of blood, as compared to intact animals; however, on the 26th day after the experiment there was a decline in concentration of blood plasma corticosterone. The hormone level dropped, as compared to intact control animals, after a radiation dose of 220 rad, both on the 1st and 26th days after the experiment.

It was previously demonstrated that there is a significant decrease in blood plasma corticosterone in rats after completion of a 22-day space experiment

on Kosmos-605 [2]. A decrease in concentration of blood plasma corticosterone in rats was also observed in the case of prolonged hypokinesia [3,4].

Corticosterone content (μg%) in rat blood plasma

| Experiment | Time after exper., day | Intact control | Experiment, rad | |
|---|---|---|---|---|
| | | | 800 | 220 |
| Flight | 1 | 10,1±1,22 | 13,2±1,42 | 18,8±3,04 |
| | *n* | 7 | 5 | 3 |
| | *P* | | >0,2 | <0,05 |
| | 26 | 18,5±1,04 | 21,1±1,23 | 14,1±2,00 |
| | *n* | 6 | 4 | 5 |
| | *P* | | >0,1 | >0,05 |
| Ground-based "control-2" | 1 | 23,6±1,81 | 26,9±3,48 | 15,7±2,15 |
| | *n* | 7 | 4 | 5 |
| | *P* | | >0,5 | <0,02 |
| | 26 | 25,6±1,58 | 17,3±1,79 | 12,5±1,03 |
| | *n* | 6 | 6 | 5 |
| | *P* | | <0,01 | <0,001 |

A comparison of the data we obtained in the Kosmos-690 experiment to the results of studies in the Kosmos-605 biosatellite experiment suggests that the combination of weightlessness and radiation (in a dosage of 220 rad) led to elevation of corticosterone concentration in blood plasma, whereas irradiation of animals on the ground elicited a decrease in blood hormone content. It should also be noted that more marked changes were observed in the case of delivery of 220 rad radiation.

Thus, these studies indicate that exposure to 800 rad γ-radiation under space flight conditions does not induce significant changes in blood plasma corticosterone content, whereas exposure to low doses (220 rad) is associated with a reliable increase in concentration of this hormone in blood.

## BIBLIOGRAPHY


1. Popens, Ya.; Silin'sh, E.; and Vitols, I. VOPR. MED. KHIMII [Problems of Medical Chemistry], No 6, 1962, pp 628-634.
2. Ilyin, E. A.; Serova, L. V.; Portugalov, V. V.; et al. AVIAT. SPACE ENVIRONM. MED., Vol 46, 1975, pp 319-321.
3. Kovalenko, Ye. A. KOSMICHESKAYA BIOL. [Space Biology], No 1, 1976, pp 3-15.
4. Fedorov, I. V. Ibid, No 4, 1971, pp 59-61.

UDC: 612.816.014.47

# EXCITABILITY OF NEUROMUSCULAR SYSTEM OF MONKEYS DURING ORTHOSTATIC TESTS

Moscow KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA in Russian
No 6, 1977 pp 79-81

[Article by G. S. Belkaniya, submitted 1 Dec 75]

[Text] The antigravity function of the organism is not only associated with increased excitability of brain structures, but alteration of functional state of the neuromuscular system which is directly involved in interaction between the body and gravity, in the form of active or passive orientation of the body in space. Information [1, 2] concerning changes in bioelectrical activity of various muscles that are functional in the positional mode is the basis for this conception. Of some interest, in this regard, are the studies of V. S. Gurfinkel' et al. [1], who demonstrated that during posture-related activity there is also a change in stretch reflex, in addition to the change in bioelectrical activity of muscles. Furthermore, what is particularly important, is that it was demonstrated that the H-reflex is also accentuated under conditions when the tendon reflex is accentuated. In the opinion of Yu. S. Yusevich [3], more intensive electrogenesis, even without postural activity, in extensor antigravity muscles, as compared to flexors, is related to the fact that gamma-efferent regulation of proprioceptors, which depends primarily on suprasegmental, including vestibular, influences, is very important to the function of muscles of the postural-tonic antigravity type. In addition, it is assumed [1] that impulsation from muscle spindles, which organizes the possibility of self-regulation of muscular activity, is important in activation of motoneurons of muscles that are posturally functional. According to the data of G. S. Ayzikov et al. [4], the efficiency of suprasegmental influences, particularly vestibular ones, on motoneurons is determined by the degree of proprioceptive afferentation and activity of spindles.

Objective evidence of changes in excitability of motoneurons under orthostatic influences would constitute a key fact, to some extent, in elucidating the role and involvement of the neuromuscular system in the functional antigravity system.

## Method

The monosynaptic H-reflex was studied in 15 Macaca rhesus monkeys. The animals were immobilized on the platform of a turntable on their abdomen. Square-wave pulses lasting 0.05-0.1 ms delivered by a Medikor electronic stimulator were used to stimulate the tibial nerve. The active spherical electrode was firmly secured in the popliteal fossa and the silent (positive) plate electrode was placed on the shaved anterior aspect of the thigh. We used a 2-channel Medikor electromyograph to record the electrical response of the soleus muscle, using tin, rectangular, plate electrodes, 5×12 mm in size applied to the skin. The distance between the electrodes usually constituted 5-8 mm.

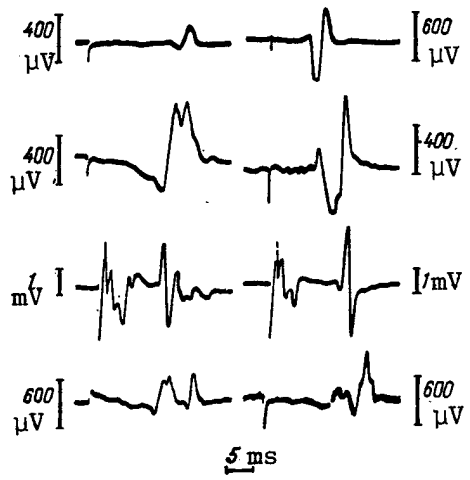



Figure 1.
Monosynaptic H-reflex in M. rhesus in horizontal position

We devoted special attention to the soleus muscle, in view of its marked postural activity, and furthermore expressly the soleus has effective monosynaptic connections with low-threshold afferent elements [5, 6]. Reflex tension of the soleus in response to stretching [7], as well as the relative magnitude of the electrical reflex response [8], are significantly greater than the response of the gastrocnemius. It is important that the M and H responses of the monosynaptic reflex are synchronized, and the number of activated motor entities can be determined from their amplitude [2, 9], so that we used the amplitude characteristics of the H response as the main test.

## Results and Discussion

The electrical response the the monkey's soleus in horizontal position had the typical form of a biphasic or triphasic potential (Figure 1), and corresponded in external characteristics to the H-reflex elicited in response to electrical stimulation in man and other animals.

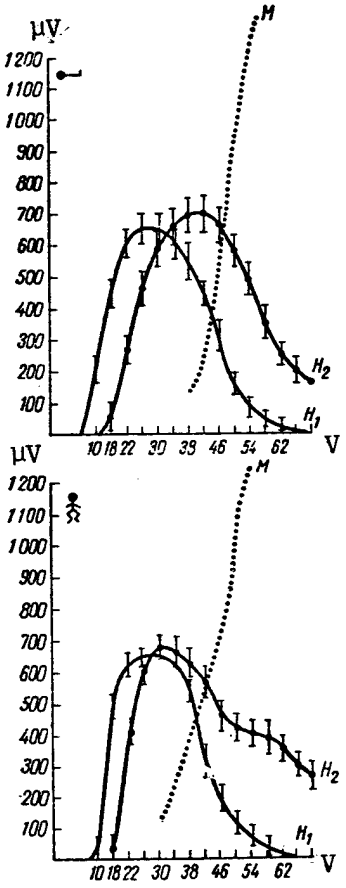



Figure 2.
Correlation between amplitude of the H response (first component, $H_1$ and second component, $H_2$) and M response, on the one hand, and force of stimulation, on the other, in horizontal and orthostatic positions (shown by the silhouettes)

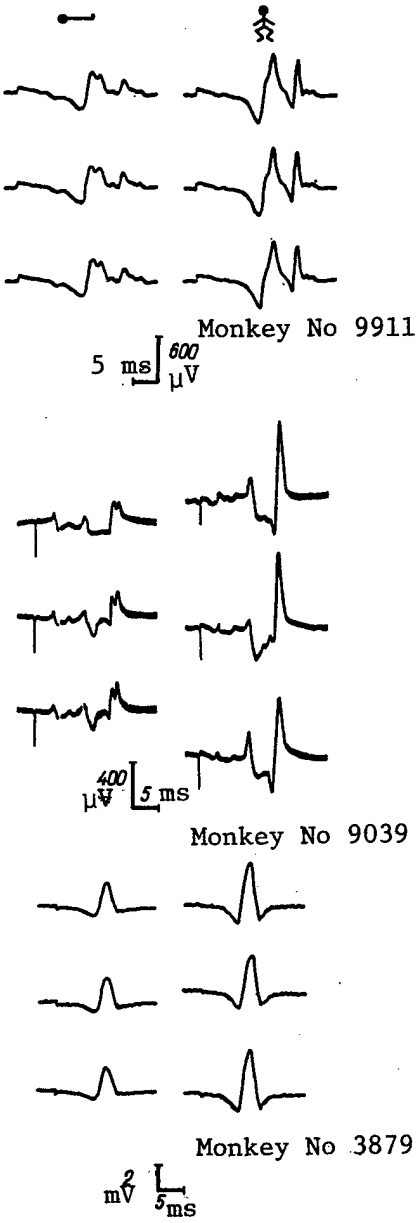



Figure 3.
Changes in H-reflex of monkeys in orthostatic test. The silhouettes illustrate the animals' position

Figure 2 illustrates a graph of amplitude of reflex response as function of force of stimulation. We can distinguish three regions on the curve of this function, which are analogous to those established for man [5]: 1) a region of rapid increase in amplitude of the H response to a maximum, with increase in force of afferent stimulus; 2) region of maximum H response,

with minor changes with continued increase in force of stimulation; 3) region of gradual decrease in amplitude of the H response. The correlation between amplitude and force of stimulation is basically the same for both components of the H response. Analysis of the correlation between amplitude of H response and force of afferent stimulus was needed, not only to confirm antidromal inhibition of motoneurons, but as a possible means of evaluating the excitability of the latter. The expedience of such an approach was confirmed in the study of the H-reflex of monkeys in the orthostatic position.

The amplitude of a response constituting 50-60% of the maximum was taken as the control level. As a rule, this corresponded to the first region of dependence of the H response on force. The choice was determined by the fact that changes in the H-reflex under the influence of different factors depend on the force of stimuli that induce them, i.e., the initial amplitude of the H-reflex [1]. In addition, it was shown [10] that the H response of only low amplitude increases under the same conditions as the tendon reflex increases. The previously existing conception that changes in tendon reflexes do not affect the mangitude of the H-reflex did not reflect the significance of initial amplitude of the monosynaptic response. In our first studies, the maximum (second region) H response showed virtually no change in orthostatic tests. This is when we took an amplitude constituting 50% of the maximum as the control amplitude of the H-reflex, and in all of the experiments we obtained a distinct increase in amplitude of the H wave ($P<0.01$)(Figure 3), which is definitely indicative of increased excitability of motoneurons in the presence of antigravity activity. This was manifested by activation of a greater number of motor units for afferent stimulation. In addition, with orthostatic orientation of the body, there is apparently some equalization (stabilization) of levels of excitability of different motoneurons, which is reflected in the steeper rise of the curve of the force characteristics of the H-reflex (see Figure 2) in the first region. This distinction is demonstrable in the dynamics of change in amplitude of both components, but especially the $H_2$ response.

Our studies revealed that the method of monosynaptic testing (H-reflex) on monkeys is quite informative. One of the real manifestations of antigravity function in them is the increase in excitability of motoneurons and number of activated motor units. Intensification of functional activity of the neuromuscular system, induced by orthostatic orientation of the body in space, is one of the chief factors providing for a certain level of background activation of central nervous structures and organization of antigravity function of the organism.

## BIBLIOGRAPHY

1. Gurfinkel', V. S.; Kots, Ya. M.; and Shik, M. L. "Regulation of Man's Posture," Moscow, 1965.

2. Person, R. S. "Electromyography in Studies of Man," Moscow, 1969.

3. Yusevich, Yu. S. "Electromyography of Tonus of Human Skeletal Muscles Under Normal and Pathological Conditions," Moscow, 1963.

4. Ayzikov, G. S.; Yemel'yanov, M. D.; Ovechkin, V. G.; et al. KOSMICHESKAYA BIOL. [Space Biology], No 3, 1971, pp 23-27.

5. Kots, Ya. M., and Krinskiy, V. I. FIZIOL. ZH. SSSR [Physiological Journal of the USSR], No 7, 1967, pp 784-790.

6. Idem, Ibid, No 1, 1968, pp 17-23.

7. Denny-Brown, D. E. PROC. ROY. SOC. LONDON, B, Vol 104, 1929, p 371.

8. Levy, R. J. NEUROL. NEUROSURG. PSYCHIATR., Vol 26, 1963, p 148.

9. Gurfinkel', V. S.; Kots, Ya. M.; Krinskiy, V. I.; et al. BYULL. EKSPER. BIOL. [Bulletin of Experimental Biology], No 5, 1965, pp 15-18.

10. Landau, W. M., and Clare, M. H. ARCH. NEUROL. (Chicago), Vol 10, 1964, p 117.

UDC: 614.777:575.76:612.46

# SANITARY AND HYGIENIC EVALUATION OF WATER REGENERATED FROM URINE

Moscow KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA in Russian
No 6, 1977 pp 82-84

[Article by N. I. Omel'yanets, L. I. Artemenko, L. P. Vlasova, N. V. Martyshchenko and S. I. Nozdrachev, submitted 3 Aug 76]

[Text] The objective of this study was to make a hygienic appraisal of water regenerated from human urine.

## Method

Regeneration was performed on an experimental device by means of indirect freezing of urine. The formed crystals of unsalted ice were separated by centrifugation. After thawing, the formed liquid was submitted to additional treatment with ion exchange resins in a mixed-action filter, in a ratio of one aliquot cation-exchange resin to 1.5 aliquots anion-exchange resin, and activated charcoal; this was then enriched with salts by filtration through solid mineralizer containing salt compounds and, finally, it was decontaminated with ionic silver.

We determined the quality of the water by studying its physicochemical properties, chemical composition and microbial content. We appraised the odor, flavor, active reaction, total hardness, levels of calcium, magnesium, chlorides, sulfates, bicarbonates, nitrogen-containing minerals, total iron, fluorine, sodium and potassium (according to difference between cations and anions), ionic silver (diphenyl thiocarbazone method), urea (by colorimetry with n-dimethylamino benzaldehyde), permanganate and biochromate oxidizability and total bacterial content [1, 2].

We conducted a chronic sanitary and toxicological experiment for 6 months to investigate the effect of this water on the organism of warm-blooded animals. This study was pursued on two groups of male mongrel rats weighing 140-165 g, with 15 animals in each group.

The first group of animals was given regenerated water daily, in automatic dispensers, and the second group was given tap water. The rats were maintained in the vivarium, on the usual diet. They were weighed monthly and weight

gain was recorded. We assessed the hemopoietic system on the basis of testing peripheral blood (hemoglobin, color index, number of erythrocytes, leukocytes and formed white blood elements) [3, 4].

Table 1. Indices of physicochemical properties of water regenerated from human urine (M±m; mean of three tests)

| Indices | Water | |
|---|---|---|
| | regenerated | tap (control) |
| Color | Clear | Clear |
| Odor at 20°, rating points | 0—1 | 0—2 |
| pH | 7,36±0,46 | 7,40±0,50 |
| Alkalinity, mg/l | 58,83±33,48 | 220,0±24,0 |
| Chlorides, mg/l | 13,24±9,22 | 9,9±8,1 |
| Sulfates, mg/l | 178,52±93,0 | 22,5±5,0 |
| Ammonia nitrog, mg/l | 2,09±0,71 | 0,2±0,05 |
| Nitrite " , mg/l | 0,032±0,036 | 0,03±0,01 |
| Nitrate " , mg/l | 0,0 | 0,3±0,1 |
| Total iron, mg/l | traces —0,04 | 0,3±0,1 |
| Overall hardness, mg-eq/l | 2,4±0,84 | 2,8±1,0 |
| Calcium, mg/l | 38,74±11,77 | 39,6±5,0 |
| Magnesium, mg/l | 5,67±3,06 | 8,5±2,1 |
| Sodium+potassium (differ.), mg-eq/l | 2,57±0,89 | — |
| Permanganate oxidizability, mg $O_2$/l | 4,63±3,04 | 3,5±0,3 |
| Bichromate oxidizability, mg $O_2$/l | 9,90±9,11 | 12,0±4,0 |
| Dry residue, mg/l | 299,0±122,54 | 320,0±86,0 |
| Ionic silver, mg/l | 0,25±0,04 | 0,0 |
| Urea, mg/l | 9,0±2,9 | 0,0 |
| Fluorine, mg/l | 0,13±0,029 | 0,10±0,02 |

To evaluate defense functions of the organism, we studied immunological reactivity according to phagocytic activity of blood leukocytes, determining the phagocyte number [4] and phagocytic index [5]. We examined various liver functions: 1) detoxification, according to prothrombin time of blood clotting [4]; 2) enzyme-synthesizing, according to levels of free sulfhydryl groups in blood serum in the course of the test, as well as after the experiment, in both hepatic and renal tissues by the method of amperometric titration [6, 7]; glycogen-synthesizing, according to glycogen content of visceral tissues using photocolorimetry with anthrone [8]. The reaction of the excretory system to ingestion of regenerated water was assessed on the basis of blood urea content assayed by photocolorimetry with diacetylmonoxime [9] and blood alkali reserve [10]. In view of the fact that the hypophyseo-adrenocortical system is capable of reacting to toxic substances, we estimated the weight coefficients of the adrenals and their ascorbic acid content by titrometry using Tilman's indicator [11]. These studies were performed monthly.

We also investigated a number of indices characterizing different physiological reactions of the organism: intratissular temperature, heart and respiration

rates. We used a PTEM-1 electric thermometer, 4EEG-3 electroencephalograph, a special sensory to determine the respiratory rate with subsequent recording of electrical marker signals on the tape of the 4EEG-3.

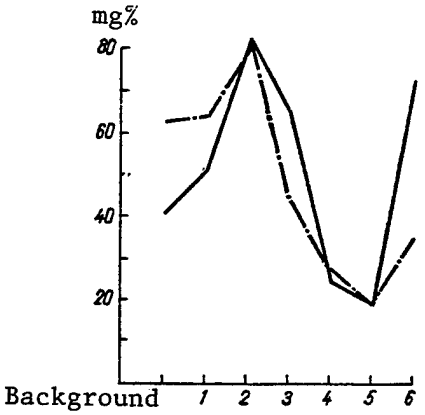
Table 2. Indices of effect of regenerated water on the organism in a chronic experiment (M±m)

| Indices | Group | | | |
|---|---|---|---|---|
| | experimental | | control | |
| | before exper. | after experiment | before exper. | after exper. |
| Weight gain, g | 161,1±2,7 | 287,5±11,5 | 157,1±3,2 | 257,1±7,0 |
| Blood hemoglobin, g% | 10,7±0,11 | 12,6±0,18 | 11,0±0,17 | 11,7±0,30 |
| Blood erythrocytes, millions/mm$^3$ | 4,8±0,1 | 6,3±0,2 | 4,6±0,1 | 5,8±0,4 |
| Blood leukocytes, thousands/mm$^3$ | 9,3±0,6 | 8,3±0,7 | 8,6±0,9 | 8,7±0,8 |
| Segmented neutrophils in blood, % | 39,5±3,8 | 31,0±4,6 | 42,1±3,3 | 28,0±3,9 |
| Blood lymphocytes, % | 54,6±3,0 | 58,5±5,0 | 47,0±2,8 | 63,1±4,0 |
| Phagocytic activity of blood leukocytes (phag. index) | 1,61±0,03 | 1,46±0,07 | 1,73±0,06 | 1,87±0,22 |
| Free sulfhydryl group content in: | | | | |
| blood serum, μmole/100 ml | 1070±9,8 | 1240±53,0 | 1070±18,5 | 1220±22,7 |
| liver tissue, μmole/100 mg | — | 1660±70,1 | — | 1680±84,2 |
| renal tissue, μmole/100 mg | — | 1680±70,2 | — | 1630±98,2 |
| Prothrombin time, s | 14,5±0,5 | 18,8±1,2 | 14,3±0,9 | 21,3±0,9 |
| Blood alkaline reserve, % | 19,2±2,4 | 38,7±3,9 | 20,7±1,5 | 37,7±3,0 |
| Intratissue temp., degrees | 36,3±0,2 | 36,2±0,2 | 36,1±0,2 | 36,3±0,2 |
| Heart rate, per min | 410±6,3 | 438±5,8 | 410±7,2 | 414±7,5 |
| Respiratory excursions/min | 112±16,3 | 126±15,0 | 112±16,4 | 94±22,3 |
| Ascorbic acid content of adrenal tissue, mg% | — | 286,5±44,9 | — | 297,3±27,1 |
| Glycogen content of hepatic tissue, mg% | — | 50,6±2,5 | — | 65,6±7,4 |
| Weight coefficients of viscera (brain, heart, liver, kidneys, adrenals) | — | No differences | — | No differences |
| Pathomorphology and histology of viscera | — | » » | — | » » |

Note: $P>0.05$.

Results and Discussion

Table 1 lists the data on the quality of regenerated water. The water was consistent with sanitary and hygienic requirements of drinking water, with respect to physical properties, cation and anion composition and total salt content. However, it had a high level of a component specific to urine, urea (9.0±2.9 mg/ℓ), although it did not exceed the maximum permissible concentration for water in reservoirs.




Blood urea content in experimental (solid line) and control (dot-dash line) animals.

X-axis, urea content; y-axis, time

Albino rats that ingested 8.0-10.0 mℓ daily of regenerated water did not differ from controls with regard to activity. According to the data listed in Table 2, there were no undesirable changes in the animals' organism. We demonstrated a brief (in the 3d and 6th months) elevation of urea level in the blood of experimental animals (see Figure). By the end of the experiment, its concentration in blood reached 0.82 g/ℓ. In view of the data indicating that urea can affect the organism only when its level in blood rises to 2.4 g/ℓ, the demonstrated elevation is most probably not deleterious [12].

Regeneration of water by means of freezing and subsequent additional treatment by the sorption method, addition of salts and decontamination with ionic silver yields water from human urine, which meets the sanitary and hygienic requirements for potable water. The obtained results are consistent with the data of S. V. Chizhov et al. [13]. On the basis of these studies, this technological system is recommended to provide water for crews of spacecraft.

BIBLIOGRAPHY

1. Kul'skiy, L. A. "Silver Water--Properties and Applications," Kiev, 1962.

2. Uspenskiy, V. A., and Gaydadymov, V. B. KOSMICHESKAYA BIOL..[Space Biology], No 1, 1972, pp 86-88.

3. Al'tgauzen, A. Ya. "Clinical Laboratory Diagnostics," Moscow, 1959.

4. Predtechenskiy, V. Ye. "Manual of Clinical Laboratory Tests," Moscow, 1960.

5. Petrov, N. S.; Nguyen Tkhe Kkhan' PROBL. GEMATOL. [Problems of Hematology], No 9, 1965, pp 33-38.

6. Gevorkyan, S. G. in: "Gigiyena i toksikologiya novykh pestitsidov i klinika otravleniy" [Hygiene and Toxicology of New Pesticides, and Symptomatology of Poisoning], Moscow, 1962, pp 321-326.

7. Pushkina, N. N. "Biochemical Methods," Moscow, 1963.

8. Morris, D. SCIENCE, Vol 107, 1948, pp 254-255.

9. Todorov, Y. "Clinical Laboratory Tests in Pediatrics," Moscow, 1963.

10. Afanas'yev, M. G. LABOR. DELO [Laboratory Record], No 4, 1969, pp 253-256.

11. Birch, T. W.; Harris, L. J.; and Ray, S. N. BIOCHEM. J., Vol 27, 1933, p 2.

12. Chizhov, S. V., et al. in: "Kosmicheskaya biologiya i aviakosmicheskaya meditsina" [Space Biology and Aerospace Medicine], Moscow--Kaluga, Vol 1, 1972, pp 232-233.

INDEX OF ARTICLES PUBLISHED IN 'KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA' [SPACE BIOLOGY AND AEROSPACE MEDICINE], VOLUME 11, NUMBERS 1-6, 1977

Moscow KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA in Russian
No 6, 1977 pp 85-89

[Index]

[Text] Editorials

Burnazyan, A. I.; Vorob'yev, Ye. I.; Gazenko, O. G.; Gurovskiy, N. N.; Nefedov, Yu. G.; Adamovich, B. A.; Yegorov, B. B.; Kovalev, Ye. Ye.; and Yegorov, A. D. "Main Stages and Prospects of Development of Space Biology and Medicine," 5(3).

Rudnyy, N. M. "Aviation Medicine on the 60th Anniversary of the Great October Revolution," 5(12).

Surveys

Alyakrinskiy, B. S. "Present Status of Space Biorhythmology," 2(3).

Vasil'yev, P. V., and Glod, G. D. "Psychopharmacology in Aviation and Cosmonautics," 3(3).

Kovalenko, Ye. A. "Principal Methods of Simulating the Biological Effects of Weightlessness," 4(3).

Popov, I. G. "Hygiene of Nutrition for Flight Personnel During Long-Term Flights," 1(3).

Sarkisov, I. Yu., and Shipov, A. A. "Biomechanical Criteria of Artificial Gravity," 5(18).

Serova, L. V. "Changes in Gravitation Level as a Stress Factor," 5(25).

Experimental and General Theoretical Research

Abrosov, N. S., and Kovrov, B. G. "Species Composition of a Mixed Culture of Microalgae as Function of Lighting and Rate of Supply of Nutrients," 1(81).

Adamovich, B. A.; Borshchenko, V. V.; Vernikov, Ya. N.; Prishchep, A. G.; and Rogatovskaya, A. P. "A Study of the Pulsed Method of Laundering Underwear," 5(80).

Andrianova, L. A., and Smirnova, N. P. "Motor Activity of Mice in Magnetic Fields Varying in Intensity," 1(54).

Andriyako, L. Ya.; Voloshin, V. G.; and Degtyarev, V. A. "Hemodynamics of Healthy Individuals as Related to Various Levels of Lower Body Negative Pressure," 1(50).

Apanasenko, Z. I. "Vestibulotonic Reflexes in Muscles of the Rat Hind Limb Following a Flight on Kosmos-605," 4(28).

Ardashnikova, L. I., and Chudnovskaya, L. A. "Rheographic Study of the Human Cardiovascular System With Exposure to High Pressure for Many Days," 2(60).

Borodkina, A. G., and Nakhil'nitskaya, Z. N. "Effect of Superhigh Intensity Constant Magnetic Fields on Morphological Composition of Peripheral Blood," 4(56).

Beregovkin, A. V.; Krupina, T. N.; Syrykh, G. D.: Korotayev, M. M.; Kuklin, N. A.; Balandin, V. A.; Znamenskiy, V. S.; Nikiforov, V. I.; Nistratov, V. V.; and Portnov, V. D. "Results of Clinical Examination of Cosmonauts After a 63-Day Flight," 2(19).

Breslav, I. S., and Salazkin, V. N. "Dynamic Control of Atmosphere Parameters in Spacecraft," 3(53).

Burkovskaya, T. Ye. "State of Hemopoiesis Under Conditions Simulating Exposure to Radiation in Long-Term Space Flights," 1(68).

Vasil'yev, V. K.; Guyev, A. A.; Savvin, A. B.; and Svirezhev, Yu. M. "Prediction and Analysis of Cosmonauts' Pulse Rate by the Method of Extrapolation Modeling in the Class of Differential Equations," 2(42).

Vasil'yev, P. V.; Belay, V. Ye.; Gaydamakin, N. A.; Glod, G. D.; Lysak, V. F.; Mel'nikova, Ye. P.; Petrukhin, S. V.; and Uglova, N. N. "Effect of Long-Term Hypokinesia on the Course of Acute Aseptic Inflammation," 4(41).

Gazo, M.; Jankela, J.; Sabo, W.; and Bodja, K. "Effects of Accelerations, Additional Weight Load and Hypokinesia on Protein Catabolism in the Japanese Quail (Coturnix Coturnix Japonica)," 1(30).

Gazenko, O. G.; Prokhonchukov, A. A.; Panikarovskiy, V. V.; Tigranyan, R. A.; Kolesnik, A. G.; Pakhomov, G. N.; Grigor'yan, A. S.; Antipova, Z. P.; Remezov, S. M.; and Komissarova, N. A. "Condition of Microscopic and Crystalline Structures, Microconsistency and Mineralization of Human Bone Tissue Following a Long Space Flight," 3(11).

Gazenko, O. G., and Shepelev, Ye. Ya. "Long-Term Space Flights and Man's Habitat," 1(10).

Gazenko, O. G.; Shumakov, V. I.; Volynkin, Yu. M.; Shal'nev, B. I.; Kakurin, L. I.; Mikhaylov, V. M.; Katkov, V. Ye.; and Chestukhin, V. V. "Technique for Selective Catheterization of the Heart and Great Vessels Used in Biomedical Studies of Healthy Individuals," 5(47).

Gerasimova, G. K., and Nakhil'nitskaya, Z. N. "Electrolyte Content of Animal Blood and Transport of Potassium Ions in Erythrocytes Under the Influence of a Constant Magnetic Field," 3(63).

Gorbunova, A. V., and Portugalov, V. V. "Cytochemical Studies of Proteins and RNA in Individual Spinal Cord Motoneurons and Spinal Ganglia Neurons of Rats Following a Space Flight," 4(24).

Grigro'yev, A. I.; Kozyrevskaya, G. I.; Dorokhova, B. R.; Lebedev, V. I.; and Morukov, B. V. "Distinctions of Fluid-Electrolyte Metabolism and Renal Function in Crew Members of the First Salyut-4 Mission," 5(41).

Grigor'yev, Yu. G.; Druzhinin, Yu. P.; Verigo, V. V.; and Il'in, Ye. A. "Main Objectives and Results of Radiobiological Experiment on Kosmos-609 Biosatellite," 5(58).

Davydov, B. I., and Antipov, V. V. "Reaction of the Organism to Hypoxia Following Gamma Irradiation," 4(55).

Degtyarev, V. A.; Doroshev, V. G.; Batenchuk-Tusko, T. V.; Kirillova, Z. A.; Lapshina, N. A.; Ponamarev, S. I.; and Ragozin, V. N. "Studies of Circulation Under Lower Body Negative Pressure on the Salyut-4 Orbital Station," 3(26).

Doroshev, V. G.; Batenchuk-Tusko, T. V.; Lapshina, N. A.; Kukushkin, Yu. A.; Kalmykova, N. D.; and Ragozin, V. N. "Changes in Hemodynamics and Phasic Structure of the Cardiac Cycle in Crew Members of the Second Salyut-4 Mission," 2(26).

Druzhinin, Yu. P.; Zubkova-Mikhaylova, Ye. I.; and Podluzhnaya, G. N. "Circadian Changes in Activity of the Hypothalamus-Hypophysis-Adrenal System in Animals Differing in Individual Radiosensitivity," 6(40).

Durnova, G. N.; Kaplanskiy, A. S.; and Portugalov, V. V. "Effect of 22-Day Space Flight on Rat Lymphoid Organs," 2(53).

Zorya, L. V. "Effect of Seduxen on the Course of Experimental Hypokinesia," 6(68).

Il'ina-Kakuyeva, Ye. I., and Portugalov, V. V. "State of Rat Muscle Motoneuron System in the Case of Restricted Mobility," 6(31).

Il'ina-Kakuyeva, Ye. I.; Portugalov, V. V.; and Krivenkova, N. P. "Effects of Space Flight Factors on Rat Skeletal Muscles," 1(20).

Ilyushko, N. A.; Il'in, Ye. A.; Kondrat'yev, Yu. I.; Korol'kov, V. I.; Kokoreva, L. V.; and Khodkevich, Yu. N. "Fluid-Electrolyte Metabolism in Albino Rats After Flight on the Kosmos-690 Biosatellite," 6(23).

Itsekhovskiy, O. G.; Polyakova, A. P.; and Lyamin, V. T. "Evaluation of Dynamics of Pulse Rate in Crew Members of Second Salyut-4 Mission at Rest and During Performance of Functional Tests in Flight," 2(37).

Kabachenko, A. N., and Fedorenko, B. S. "Cataractogenic Effects of 25 and 50 MeV Protons," 4(59).

Kazaryan, V. A.; Rapoport, E. A.; Goncharova, L. A.; and Bulycheva, S. Ya. "Effects of Prolonged Weightlessness on Metabolism of Proteins in Red and White Skeletal Muscles of Rats," 6(19).

Kalinichenko, V. V. "Dynamics of Orthostatic Stability of Cosmonauts After Flights Lasting 2 to 63 Days," 3(31).

Katkov, V. Ye.; Kauricheva, N. I.; Chestukhin, V. V.; Zybin, O. Kh.; Seyfulla, R. D.; and Utkin, V. N. "Indices of Acid-Base Equilibrium and Enzymatic Activity of Human Blood During Brief Antiorthostatic Hypokinesia," 5(51).

Kovalenko, Ye. A.; Popkov, V. L.; Lakota, N. G.; and Kantor, S. L. "Patterns of Changes in the Canine Organism With Malfunction of Atmosphere Regeneration System in a Sealed Tank," 1(58).

Kozlova, V. G.; Oganov, V. S.; and Rakhmanov, A. S. "Investigation of Mechanical Activity of Canine Skeletal Muscles in Vivo," 2(79).

Kolemeyeva, L. Ya.; Shashkov, V. S.; and Yegorov, B. B. "Animal Sensitivity to Central Nervous System Stimulators During Hypokinesia," 2(74).

Komolova, G. S.; Guseynov, F. T.; Makeyeva, V. F.; Yegorov, I. A.; Tigranyan, R. A.; and Serova, L. V. "State of DNA in Rat Liver and Spleen After Space Flight in Kosmos-605," 4(17).

Korotayev, M. M.; Popov, I. I.; Degtyarev, V. A.; Dorofeyeva, Z. Z.; Yegorov, A. D.; Kalinichenko, V. V.; Ponomarev, S. I.; Sidorov, V. P.; Polyakova, A. P.; Golubchikova, Z. A.; and Savilov, A. A. "Electrocardiographic Examination of Crew of Second Mission in Salyut-4," 2(22).

Pak, Z. P.; Koloskova, Yu. S.; and Chizhov, S. V. "Hygienic Evaluation of Water Regenerated from Diverse Moisture-Containing Waste," 6(57).

Pestov, I. D.; Asanov, Kh. Kh.; Asyamolov, B. F.; Karpusheva, V. A.; Kas'yan, I. I.; Panchenko, V. S.; and Stepantsov, V. I. "Experimental Evaluation of Relationship Between Pulmonary Blood Flow, Fluid-Electrolyte Metabolism and Orthostatic Reactions of Man," 2(68).

Petrova, N. V., and Portugalov, V. V. "Lactate Dehydrogenase Isozymes of Rat Skeletal Muscles After Space Flight and During Hypokinesia," 5(66).

Petrovykh, V. A.; Shovkoplyas, O. A.; Mikhel'son, D. A.; and Aronova, Ye. N. "Indices of Nitrogen, Carbohydrate and Lipid Metabolism in Man as Related to Prolonged Exposure to Hyperbaric Conditions," 1(48).

Pilipyuk, Z. I.; Gorban', G. M.; Solomin, G. I.; and Gorshunova, A. I. "Toxicology of 1-4-Dioxane," 6(53).

Polyakov, B. I.; Petrenko, B. Ye.; Savvin, A. B.; Tazetdinov, I. G.; and Tarko, A. M. "Cardiovascular Reactions to Orthostatic Tests and Human Resistance to Vestibular Stimuli," 3(48).

Polyakov, V. V.; Agureyev, A. N.; Vlasova, T. F.; and Ushakov, A. S. "Some Indices of Phosphocreatine Metabolism in Man as Related to High and Low Levels of Energy Expenditure," 1(43).

Potapov, P. P. "Tissue Mucopolysaccharides and Collagen in Hypokinetic Rats," 3(44).

Prokhonchukov, A. A.; Tigranyan, R. A.; Kolesnik, A. G.; Novikov, L. L.; Timofeyeva, N. T.; Zhizhina, N. A,; Volozhin, A. I.; Neustroyev, G. V.; and Matviyenko, V. P. "Calcium and Phosphorus Content, and Incorporation of Calcium 45 in Bones and Teeth of Rats Following 22-Day Orbital Flight in Kosmos-605 Satellite," 1(26).

Radkevich, L. A. "Model of Motion Sickness in Dogs Used to Evaluate Efficacy of Pharmacological Agents," 6(50).

Raytses, I. V. "Effect of Electric Stimulation of the Hypothalamus and Limbic Structures on Vestibulosomatic Reflexes," 1(78).

Rapoport, E. A.; Goncharova, L. A.; Morenkova, S. A.; and Kazaryan, V. A. "Effect of Long-Term Space Flight on Biosynthesis of Proteins in Different Rat Organs and Tissues," 4(20).

Rerberg, M. S.; Manukovskiy, N. S.; and Somova, L. A. "Intestinal Autoflora of Test Subjects in the Course of a 6-Month Bioengineering Experiment," 2(57).

Rudnyy, N. M.; Gazenko, O. G.; Gozulov, S. A.; Pestov, I. D.; Vasil'yev, P. V.; Yeremin, A. V.; Degtyarev, V. A.; Balakhovskiy, I. S.; Bayevskiy, R. M.; and Syrykh, G. D. "Principal Results of Medical Research Pursued During the Flights of Two Crews on Salyut-5 Orbital Station," 5(33).

Savina, Ye. A.; Pankova, A. S.; and Alekseyev, Ye. I. "Morphological Manifestations of Functional Changes in the Hypothalamohypophyseal Neurosecretory System and Kidneys of Rats Under the Influence of Space Flight Factors," 5(72).

Simonov, Ye. Ye. "Acute Leukocytic Reactions to Impact Accelerations," 4(62).

Simonov, Ye. Ye., and Ryklin, K. B. "Functional State of the Rat Liver Under the Primary Deleterious Effect of Impact Accelerations," 6(45).

Stazhadze, L. L.; Bogomolov, V. V.; and Goncharov, I. B. "Problems of Medical Back-Up of the Immediate Postflight Period Following Long-Term Space Flights," 4(14).

Tigranyan, R. A.; Popova, I. A.; Belyakova, M. I.; Kalita, N. F.; Tuzova, Ye. G.; Sochilina, L. B.; and Davydova, N. A. "Results of Studying Metabolic Processes in Crew Members of the Second Salyut-4 Orbital Station Mission," 2(48).

Tikhomirov, I. B. [deceased]; Turchaninova, V. F.; Selivanenko, V. T.; and Staferov, V. A. "Use of Tetrapolar Rheography to Evaluate the Circulatory System," 4(70).

Tikhonov, M. A., and Belan, A. S. "Mechanics of Speech-Related Respiration While Performing Static Physical Exercise," 3(58).

Chekirda, I. F., and Yeremin, A. V. "Dynamics of Cyclic and Acyclic Locomotion of the Soyuz-18 Crew After a 63-Day Space Mission," 4(9).

Cherepakhin, M. A.; Kakurin, L. I.; Il'ina-Kakuyeva, Ye. I.; and Fedorenko, G. T. "Evaluation of Efficacy of Electric Stimulation of Muscles in Preventing Disorders Related to Prolonged Restriction of Motor Activity in Man," 2(64).

Chernyakov, I. N.; Maksimov, I. V.; and Glazkova, V. A. "Prevention of Decompression Sickness Related to Brief High-Altitude Flights in Depressurized Aircraft Cockpits," 1(63).

Shabel'nikov, V. G. "Variants of Control for an Ecosystem That is Closed With Regard to Exchange of Gases, With Periodically Functioning Autotrophic Component," 6(63).

Shubich, M. G.; Goryacheva, L. L.; Dudetskiy, V. I.; Lutsenko, N. M.; and Mogil'naya, G. M. "Histochemical Studies of Rat Digestive Organs After Flight on Kosmos-605," 3(40).

AUTHOR INDEX

Moscow KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA in Russian
No 6, 1977 pp 89-91]

[Author index]

ANNOUNCEMENT CONCERNING BOOK ON MICROCHEMICAL ANALYSIS OF NERVOUS TISSUE

Moscow KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA in Russian
No 6, 1977 inside back cover

[Announcement for readers]

[Text] "Microchemical Analysis of Nervous Tissue," by N. N. Osborne, originally published in England, in 1974, has been published by Meditsina Publishing House, as part of the 1978 plan for the publication of literature. This is a translation from English, published in 1978 (first quarter), on 20 sheets; it is illustrated; the printing is 3000 copies and price is 3.36 rubles.

This book deals with methods of microbiochemical demonstration in isolated neurons of the animal brain of various fractions of protein, amines, amino acids, phospholipids, nucleic acids, some enzymes, mediators, as well as ions. It describes methods of isolating neurons, incubation thereof, weighing and homogenization, as well as information about biochemical heterogeneity of neurons in various parts of the animal brain and changes in metabolism thereof under the influence of some factors used in vivo and in vitro.

This book will be of interest to biochemists.

It was published under the plan for 1978, No 339.

Books published by Meditsina Publishing House are sold in specialized book stores and stores with departments of medical literature.

ANNOUNCEMENT FOR PHYSICIANS CONCERNING GENTAMYCIN SULFATE

Moscow KOSMICHESKAYA BIOLOGIYA I AVIAKOSMICHESKAYA MEDITSINA in Russian
No 6, 1977 back cover

[Announcement issued by the All-Union Marketing and Information Office, Main Pharmaceutical Administration, USSR Ministry of Health]

Text] Gentamycin Sulfate (Gentamycin sulfas), in powder form, dispensed in vials, 0.08 g in each.

Synonyms: garamycin, garimycin, ribofacin, etc.

Gentamycin sulfate has a wide spectrum of antimicrobial action. It is active with regard to most Gram-negative and Gram-positive microorganisms.

It is rapidly absorbed when injected intramuscularly. The maximum concentration of this product in blood is demonstrable 1 h after administration; therapeutic concentrations are retained for 8 h. The concentration of this antibiotic in blood increases in patients with impaired renal function.

Gentamycin sulfate is used for various infectious diseases induced by microorganisms that are sensitive to this drug (pneumonia, bronchopneumonia, pleurisy, lung abscess, sepsis, meningitis, peritonitis, malignant endocarditis, etc.).

This product is effective in cases of urinary tract infections (pyelonephritis, cystitis, urethritis), infected burns, as well as suppurative septic pathology in patients with leukemia and malignant neoplasms, against the background of cytostatic, radiation therapy and administration of immunosuppressants. Gentamycin sulfate can be prescribed prior to isolation of a pathogen and determination of its sensitivity to this antibiotic.

Gentamycin sulfate is administered intramuscularly.

For urinary tract infections, the single dosage for adults is 0.4 mg/kg, and the daily dosage is 0.8-1.2 mg/kg (given in 2-3 divided doses). The mean course of therapy lasts 6-7 days. In the case of severe course of the infectious process, the daily dosage can be increased to 3 mg/kg, provided the concentration of this antibiotic in blood, renal functions and audiometry are monitored.

For infections localized elsewhere (peritonitis, meningitis, sepsis, etc.) the daily dosage of gentamycin sulfate may be 2.4-3.2 mg/kg or more (up to 5 mg/kg). The drug is given 2-3 times a day. Treatment is continued for 7-10 days. If necessary (depending on vital signs), the courses can be repeated after a 7-10-day interval.

Gentamycin sulfate is contraindicated in the presence of allergy to this product, uremia, pathology of the auditory and vestibular systems related to neuritis of the 8th pair of cranial nerves, and myasthenia.

Gentamycin sulfate should not be prescribed for pregnant women, unless there are vital indications.

Gentamycin sulfate cannot be given concurrently or alternately with other ototoxic and nephrotoxic products (streptomycin, kanamycin, neomycin, monomycin, ristomycin, cephaloridine, furosemide).



10,657
CSO: 1849

- END -